UNIVERSIDADE FEDERAL DE MINAS GERAIS
Escola de Veterinária
Programa de Pós-Graduação em Zootecnia

Edilane Costa Martins

Desempenho, qualidade da carne e emissões de gases de efeito estufa de bovinos zebuínos e cruzados alimentados com diferentes proporções de concentrado em confinamento

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Tese apresentada ao Programa de Pós-Graduação em Zootecnia da Escola de Veterinária da Universidade Federal de Minas Gerais como requisito parcial para obtenção do título de Doutor em Zootecnia.

Área de concentração: Produção e Nutrição Animal

Prof.ª. Orientadora: Ângela Maria Quintão Lana
Prof. Coorientador: Leandro Sâmia Lopes e
Ramon Costa Alvarenga

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ATA DE DEFESA DE TESE DA ALUNA EDILANE COSTA MARTINS

Às 14:00 horas do dia 27 de fevereiro de 2023, reuniu-se, a Comissão Examinadora de Tese, aprovada em reunião ordinária no dia 03/10/2022, para julgar, em exame final, a defesa da tese intitulada: Desempenho, qualidade da carne e emissões de gases de efeito estufa de bovinos zebuínos e cruzados alimentados com diferentes proporções de concentrado em confinamento., como requisito final para a obtenção do Grau de Doutor em **Zootecnia, área de concentração Produção de Ruminantes.** Abrindo a sessão, o Presidente da Comissão, Profa. **Ângela Maria Quintão Lana**, após dar a conhecer aos presentes o teor das Normas Regulamentares da Defesa de Tese, passou a palavra ao (a) candidato (a), para apresentação de seu trabalho. Seguiu-se a argüição pelos examinadores, com a respectiva defesa do candidato (a). Logo após, a Comissão se reuniu, sem a presença do candidato e do público, para julgamento da tese, tendo sido atribuídas as seguintes indicações:

	Aprovada	Reprovada
Prof.(a)/Dr.(a) Bruno José Rodrigues Alves	X	
Prof.(a)/Dr.(a) Isabella Cristina de Faria Maciel	X	
Prof.(a)/Dr.(a) Henrilly Cristina Menezes de Sá	X	
Prof.(a)/Dr.(a) Luciano Soares de Lima		
Prof.(a)/Dr.(a) Ângela Maria Quintão Lana	X	

Pelas indicações, o (a) candidato (a) foi considerado (a): [X] Aprovado (a) [ ] Reprovado (a)

Para concluir o Doutorado, o(a) candidato(a) deverá entregar 03 volumes encadernados da versão final da tese acatando, se houver, as modificações sugeridas pela banca, e a comprovação de submissão de pelo menos um artigo científico em periódico recomendado pelo Colegiado dos Cursos. Para tanto terá o prazo máximo de 60 dias a contar da data defesa.

O resultado final, foi comunicado publicamente ao (a) candidato (a) pelo Presidente da Comissão. Nada mais havendo a tratar, o Presidente encerrou a reunião e lavrou a presente ata, que será assinada por todos os membros participantes da Comissão Examinadora e encaminhada juntamente com um exemplar da tese apresentada para defesa.

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“...o que a vida quer da gente é CORAGEM...”

(Guimarães Rosa – Grande sertão: veredas)

RESUMO

O cruzamento de bovinos *Bos taurus taurus* x *Bos taurus indicus* se tornou uma valiosa alternativa na busca de melhora do desempenho e qualidade da carcaça e carne dos animais. Acrescenta-se ainda, que o sistema de produção adotado e as dietas fornecidas podem melhorar não só a performance animal, mas também gerar um produto com características mais saudáveis à saúde humana. O sistema intensivo de criação de bovinos de corte em confinamento tem se expandido cada vez mais pelos trópicos, mas o conhecimento a respeito do impacto que este tipo de sistema pode causar ao meio ambiente nas regiões de clima tropical ainda requer muita investigação. Nesse sentido, objetivou-se avaliar o consumo, desempenho e a qualidade da carne de bovinos Nelore e cruzados (Nelore x Angus) alimentados com dietas contendo dois níveis de concentrado (65 e 85 %), além de avaliar os fluxos de óxido nitroso (N_2O) e metano (CH_4) emitidos das excretas desses bovinos depositadas em solo de confinamento durante o período seco do ano no Brasil. Para avaliar o impacto do genótipo e da dieta sobre o consumo, o desempenho e a qualidade da carne foram utilizados, 23 Nelores com peso vivo inicial (PVI) de 377,17 kg ± 4,37 kg e 25 cruzados com PI de 415,33 kg ± 4,20 kg. Os animais foram terminados em confinamento por 84 dias e receberam dietas com dois níveis de concentrado na matéria seca (85 e 65 %). Para a avaliação do consumo de matéria seca (CMS) usando dióxido de titânio como marcador externo, foram aleatoriamente selecionados 16 animais Nelore (oito consumindo a dieta 65 % e oito consumindo a dieta 85 %) e 16 animais cruzados (oito consumindo a dieta 65 % e oito consumindo a dieta 85 %). Também foram avaliadas as variáveis de desempenho como peso vivo médio (PVM), ganho de peso (GP), ganho médio diário (GMD), peso de carcaça quente (PCQ), ganho médio diário em carcaça (GMDc), eficiência e conversão alimentar (EA, CA). Foram avaliadas as variáveis de qualidade de carne (cor, maciez, composição centesimal e perfil de ácidos graxos) e a emissão de N_2O e CH_4 das excretas depositadas no solo. Para a avaliação das variáveis de qualidade de carne os mesmos animais selecionados para a avaliação do CMS foram utilizados. Para a avaliação da emissão de N_2O e CH_4 foram utilizadas excretas frescas (Fezes e urina) de bovinos Nelore e cruzados. Foi feito um pool da urina e das fezes frescas e aplicado ao solo do confinamento em câmaras estáticas instaladas no solo de um curral de confinamento a céu aberto previamente reservado. Os fluxos de emissão de N_2O e CH_4 foram monitorados por 84 dias. Os animais cruzados apresentaram maior PVM, GMD, PCQ e GMDc. Não houve diferença estatística ($P>0,05$) entre genótipo e dieta para o CMS, porém os animais cruzados demonstraram ser mais eficientes, uma vez que apresentaram melhor desempenho consumindo a mesma quantidade de alimento que os animais Nelore. Quanto a qualidade da carne, ambos genótipos apresentaram carne com cor clara em comparação ao reportado na literatura. Não foi observada interação significativa ($P>0,05$) entre genótipo e dieta na concentração dos ácidos graxos saturados, monoinsaturados e polinsaturados. No entanto interação significativa ($P<0,05$) foi observada entre genótipo e dieta no somatório dos ácidos graxos insaturados, em que os animais cruzados alimentados com a dieta contendo 85% de concentrado apresentaram maior quantidade de ácidos graxos insaturados. Se tratando das emissões de gases de efeito estufa (GEEs) os animais cruzados apresentaram emissões de CH_4 2,4 vezes maior que os animais Nelore ($P<0,039$) e os animais que consumiram a dieta com baixo nível nutricional apresentaram 50,1% menos emissão de N_2O pelas excretas em comparação com os animais consumindo a dieta com alto nível nutricional. A emissão por CMS (E/CMS) e a emissão por GMD (E/GMD) de CH_4 foi maior nos animais cruzados em comparação com os animais Nelore. Já a E/GMD e E/CMS de N_2O dos animais consumindo a dieta com alto nível nutricional foi 18,2% e 36,1% maior para os animais cruzados comparado com os animais Nelore, respectivamente. Os resultados mostram que os animais cruzados apresentaram melhor performance em comparação aos animais Nelore e que ambos os genótipos tiveram a coloração da carne classificada como clara em comparação aos relatado na literatura. Ambos os genótipos e dietas mostraram capacidade de alteração do perfil de ácidos graxos da carne, sendo que os animais cruzados apresentaram maior quantidade de ácidos graxos insaturados em comparação aos animais Nelore. A dieta com 85% de concentrado proporcionou maior quantidade de ácidos graxos insaturados, maior relação n-6/n-3 e maior índice de trombogenicidade. Apesar do receio envolvendo o consumo de carne por causa de potenciais riscos à saúde, a carne dos animais Nelore e cruzados apresentaram grande quantidade de ácidos graxos em seu perfil que são benéficos à saúde humana. E em se tratando das emissões dos GEEs, os animais cruzados consumindo dietas com mais concentrado emitem mais CH_4 e N_2O em suas excretas. Os dados desde estudo indicam que o rebanho de animais Nelore, que é predominante no Brasil, emite menos GEEs do que outros genótipos e dietas com inclusão de alto nível de concentrado tem maior potencial de emissão de GEEs.

Palavras-chave: metano; óxido nitroso; perfil de ácidos graxos; produção sustentável.

ABSTRACT

Bos taurus indicus and *Bos taurus taurus* crossbreed has become a valuable alternative on the search to improve performance, meat, and carcass quality. It's added that the production system adopted, and the diets provided can improve not only animal performance, but also to contribute with a product with healthier characteristics for human healthy. The intensive system of beef cattle production in feedlot has been expanding more and more throughout the tropics, but the impact that this type of system can cause to the environment in tropical regions is uncertain and still requires a lot of investigation. In this sense, we aimed evaluate the intake, performance and meat quality of Nellore and crossbred animals fed diets with two levels of concentrate (85 and 65 %), in addition to evaluating the nitrous oxide (N_2O) and methane (CH_4) fluxes emitted from these cattle excreta deposited in feedlot soil used in Brazil. To evaluate the impact of genotype and diet on feed intake, performance, and meat quality 23 Nellore with IBW of 377.17 kg ± 4.37 kg and 25 crossbreed with 415.33 kg ± 4.20 kg of IBW were used. The animals were finished in feedlot for 84 days fed with two levels of concentrate in dry matter basis (85 and 65 %). To evaluate the dry matter intake (DMI) using titanium dioxide (TiO_2) as an external marker 16 Nellore (eight consuming the 85 % diet and eight consuming the 65 %) and 16 crossbreed animals (eight consuming the 85 % diet and eight consuming the 65 %) were randomly selected. The performance variables body weight (BW), gain weight (GW), average daily gain (ADG), hot carcass weight (HCW), average daily carcass gain (ADGc), feed efficiency and conversion (FE, FC), meat quality variables (color, tenderness, centesimal composition, and fatty acid profile) and N_2O and CH_4 emission from cattle excreta deposited in soil were evaluated. To evaluating the meat quality variables the same animals selected for the DMI evaluation were used. For the evaluation of N_2O and CH_4 emissions, fresh excreta (feces and urine) from Nellore and crossbreed cattle were used. Fresh urine and feces were mixed and applied together on the feedlot soil in static chambers installed on the ground of a previously reserved open-air feedlot pen. N_2O and CH_4 fluxes were monitored for 84 days. The crossbred animals showed greater BW, ADG, HCW and ADGc. There was no difference between breed or diet ($P>0.05$) in DMI. Both breeds presented lighter color meat compared to the literature. No interaction was observed ($P>0.05$) between breed or diet in the concentration of saturated, monounsaturated and polyunsaturated fatty acids. Significant interaction was observed ($P<0.05$) between breed and diet in the sum of unsaturated fatty acids, where crossbred animals fed 85% diet showed greater amount of unsaturated fatty acids. Related to greenhouse gas emissions (GHGs), CH_4 emissions were 2.4 times higher in crossbred than in Nellore animals ($P<0.039$). N2O emissions from excreta were 50.1% lower in animals consuming diet with low compared to high nutritional level. The emission per DMI (E/DMI) and emission per ADG (E/ADG) of CH_4 were higher in crossbred compared to Nellore animals, and the E/ADG and E/DMI of N_2O of animals fed high nutritional level were 18.2% and 36.1% higher in crossbred compared to Nellore, respectively. The results showed that crossbred animals had a better performance compared to Nellore and both breeds presented a lighter color meat in comparison to the literature. Both breeds and diets showed the ability to change the meat fatty acid profile. Crossbred animals showed higher concentration of unsaturated fatty acid on LT muscle compared to Nellore animals. The diet with 85% concentrate provided a greater amount of unsaturated fatty acid, n-6/n-3 ratio and thrombogenicity index. Despite the concern involving the consumption of beef because of the potential health problems, the fatty acids profile in the LT muscle showed greater amounts of fatty acids that are beneficial to human health. Related to the GHGs emission, crossbred animals consuming diets with more concentrate feed emit more CH_4 and N_2O in their excreta. These data indicate that the Nellore herd, which id predominant in Brazil, emits lower GHGs than other breeds, and that diets with high concentrate feed inclusion have greater potential to emit GHGs.

Key words: fatty acid profile; methane; nitrous oxide; sustainable production.

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LISTA DE SIGLAS E ABREVIATURAS

AGI – Ácidos graxos insaturados

AGM – Ácidos graxos monoinsaturados

AGP – Ácidos graxos poli-insaturados

AGS – Ácidos graxos saturados

C - Carbono

CA – Conversão alimentar

CH_4 – Metano

CLA – Ácido graxo linoleico conjugado

CMS – Consumo de matéria seca

CO_2 – Dióxido de carbono

DAA – Dias após aplicação

DFDN – Digestibilidade da fibra insolúvel em detergente neutro

DMO – Digestibilidade da matéria orgânica

DMS – Digestibilidade da matéria seca

DPB – Digestibilidade da proteína bruta

EA – Eficiência alimentar

EE – Extrato etéreo

E/ADG – Emissions per average daily gain

E/GMD – Emissões por ganho médio diário

E/DMI – Emissions per dry matter intake

E/CMS – Emissões por consumo de matéria seca

FC – Força de cisalhamento

FDA – Fibra insolúvel em detergente ácido

FDN – Fibra insolúvel em detergente neutro

FDNi – Fibra em detergente neutro indigestível

GEE – Gases de efeito estufa

GMD – Ganho médio diário

GMDc – Ganho médio diário em carcaça

GP – Ganho de peso

GT – Ganho total

ha – hectare

HDL – Lipoproteína de alta densidade

IMA – Instituto Mineiro de Agropecuária

LDL – Lipoproteína de baixa densidade

MM – Matéria mineral

MO – Matéria orgânica

MS – Matéria seca

N - Nitrogênio

N_2O – Óxido nitroso

NDT – Nutrientes digestíveis totais

NH_3 – Amônia

NH_4^+ - Amônio

NO_3^- - Nitrato

PB – Proteína bruta

PC – Perda por cocção

PC – Peso de carcaça

PCQ – Peso de carcaça quente

PIB – Produto interno bruto

PVF – Peso vivo final

PVI – Peso vivo inicial

PVM – Peso vivo médio

RC – Rendimento de carcaça

TiO_2 – Dióxido de titânio

UA – Unidade animal

SUMÁRIO

CAPÍTULO 1 – REVISÃO DE LITERATURA

1. Introdução geral

Os avanços na pecuária brasileira vêm acontecendo de forma gradativa, porém não imperceptível. Durante muitos anos a produção a pasto era predominantemente praticada, entretanto tem se observado crescente uso do sistema de produção intensivo, o confinamento, para terminação de bovinos. No entanto ainda há muitos desafios na produção animal, um deles é a produção de alimentos em quantidade e qualidade suficientes para suprir o aumento da população e, ao mesmo tempo, minimizar os impactos ambientais (Duthie et al., 2017). Espera-se que a população mundial aumente para mais de 9,5 bilhões de habitantes até o ano de 2050 (FAO, 2009). O crescimento populacional e mudanças no padrão de consumo quanto à exigência por uma produção animal com menor impacto ambiental resultará em aumento na demanda por alimentos que tenham processos de produção que respeitem o meio ambiente.

Com cerca de 218 milhões de cabeças em seu rebanho bovino (IBGE, 2020) a pecuária brasileira tem grande importância no cenário econômico nacional, tendo o agronegócio sido responsável por 27% do Produto Interno Bruto (PIB) em 2020. De janeiro a setembro de 2021 o agronegócio apresentou um crescimento modesto (0,4%), no entanto o ramo pecuário apresentou queda de 4,76% em relação ao mesmo período de 2020 (CEPEA/CNA, 2021). Sabendo da grande importância do setor pecuário para o país, aliado a necessidade de se aumentar e melhorar o produto produzido, além dos avanços no sistema de produção animal o investimento em melhoramento do rebanho vem sendo realizado com o intuito de obter animais que sejam superiores em desempenho. O cruzamento entre animais zebuínos e taurinos tem contribuído fortemente com a melhoria dos animais produzidos em território nacional. No entanto, apesar da contribuição positiva que a cadeia da carne e do leite traz para o país, tanto econômico quanto social, haja visto que muitas famílias retiram sua renda da pecuária, também há a contribuição negativa e neste ponto precisa-se falar sobre a emissão de gases de efeito estufa (GEE).

As emissões de GEE pela atividade pecuária representam 10-12% das emissões globais (IPCC, 2019), dos quais 6,4% correspondem às emissões de metano (CH_4) e 4,2% às emissões de óxido nitroso (N_2O) (Gerber et al., 2013). Embora não seja um valor tão expressivo, o sistema de produção de carne tem sido apontado como fonte significativa de emissões de GEE, uma vez que o potencial de aquecimento global desses gases é 25 e 298 vezes maior que o CO_2, respectivamente (IPCC, 2019).

A preocupação com a conservação do meio ambiente tem crescido cada vez mais, já que vários problemas decorrentes do mau uso dos recursos ambientais têm surgido deixando consumidores e produtores em situação de alerta. A produção de carne bovina brasileira ocupa uma posição de destaque no cenário mundial, a qual responde por 14,9% da produção (FAO, 2021). Encontrar o equilíbrio entre o aumento na produção de alimentos e a redução dos impactos ambientais, mediante a produção animal sustentável, se faz necessário. Assim a busca pela melhoria na eficiência produtiva com a utilização de práticas modernas de gestão e de tecnologias pode favorecer a produção de carne bovina de forma mais sustentável, reduzindo o impacto ao ambiente.

Neste contexto, objetivou-se com esta pesquisa avaliar o desempenho e a qualidade da carne de bovinos zebuínos e cruzados terminados em confinamento alimentados com dietas com dois níveis de concentrado e, mensurar as emissões de óxido nitroso e metano provenientes da deposição no solo de excretas de bovinos em confinamento.

1.1. Sistema de produção de bovinos de corte no Brasil

Com o maior rebanho comercial do mundo (218 milhões de cabeças), liderando as exportações mundiais de carne e com uma grande importância econômica para o Brasil a bovinocultura de corte brasileira ainda apresenta grandes entraves como taxas produtivas abaixo de suas potencialidades. Como exemplo podemos citar a taxa de lotação menor que 1 unidade animal/hectare (UA/ha) e produtividade menor que 120 kg de peso vivo ou 4 arrobas/ha/ano (IBGE, 2017). Segundo a ABIEC (2021), em 2020 a pecuária contribuiu com 10% do produto interno bruto (PIB) total brasileiro. A pecuária de corte brasileira ainda tem o pasto como seu principal sistema de produção, ocupando cerca de 163 milhões de hectares de pastagens (ABIEC, 2022), sendo então, a maioria dos bovinos de corte criados, recriados e terminados em pastagens no Brasil (Silva et al., 2016).

Atualmente é grande a preocupação com o meio ambiente, com a redução dos recursos naturais, com as mudanças climáticas e, por consequência, tem havido questionamentos sobre a intensificação das práticas de produção de carne bovina. Inclusive o mercado internacional tem demonstrado preocupação com a abertura de novas áreas de pastagens, chamada de intensificação horizontal, que resulta em desmatamentos e desflorestamentos, inadmissíveis atualmente, devido ao grande impacto causado ao ambiente. Entretanto, o estudo de Silva et al.

(2016) mostrou que apenas 10% do aumento de produção de bovinos ocorre devido à expansão de pastagens e que os outros 90% é resultante de ganhos em produtividade, principalmente devido a melhorias nas pastagens já existentes e a utilização de confinamentos e de animais cruzados com genótipos mais favoráveis a maior produção.

Nos últimos anos tem havido uma pressão crescente para que não haja mais desmatamento, concomitantemente com a mitigação de GEE alertando para uma mudança nos processos da produção animal. A intensificação vertical, com aumento da produção de carne em menor tempo e área, através de estratégias de inovação tecnológica que permitam aumentar a produção e ao mesmo tempo reduzir os prejuízos ao ambiente tem sido prioridade. Dentre as estratégias que podem ser utilizadas para intensificação vertical estão o manejo de pastagem, a suplementação energético-proteico-mineral para animais em pasto, tanto no período das águas quanto no período da seca, a integração lavoura-pecuária, o confinamento, além dos programas de melhoramento genéticos, como por exemplo, os cruzamentos entre raças de animais mais precoces (*Bos taurus taurus*) com raças de animais mais tardios (*Bos taurus indicus*). Todas essas alternativas podem contribuir de forma positiva gerando maior produtividade do sistema.

A intensificação da produção pecuária impacta na quantidade de carne produzida por área (kg/ha/ano) e nas emissões de GEE. Os sistemas de produção intensivos, como o sistema de terminação de animais em confinamento com alto grão, demandam menor área para produção e reduzem as emissões de GEE por quilograma de carne comparado aos sistemas extensivos (Swain et al., 2018), além de liberar a pastagem para categorias animais que apresentam menor demanda de mantença.

Em 2015 apenas 13% dos animais abatidos tinham sido terminados em confinamento (ABIEC, 2019). Seis anos depois 17,19% dos animais abatidos no país foram terminados em confinamento, mostrando o quanto a pecuária tem modificado a forma de produção, aumentando o número de animais confinados em busca de reduzir a idade ao abate, aumentar o peso da carcaça, e reduzir a emissão de GEE (ABIEC, 2022).

Além da transição de sistemas extensivos para intensivos de produção, o aumento da produtividade e a redução do ciclo de produção podem ser alcançados com a adoção de programas de melhoramento genético animal. Independente da intensidade e do sistema de produção utilizado, o genótipo zebuíno é predominante nos rebanhos brasileiros, principalmente animais da raça Nelore. Porém nas últimas décadas, com o intuito de melhorar a produtividade, animais de raças taurinas passaram a ser utilizados em cruzamentos nos programas de melhoramento genético, com destaque para as raças Aberdeen Angus, Charolês, Hereford e Simental.

1.2. Cruzamento industrial

A maior parte do rebanho bovino brasileiro é constituído por animais da raça Nelore, isto é devido as suas características adaptativas ao clima brasileiro, além de serem animais que apresentam, em geral, estado sadio e vigoroso. Porém, devido a vários fatores, entre eles o sistema de criação adotado pelos produtores, esses animais apresentam baixa produtividade, muitas vezes abaixo da capacidade genotípica do animal.

A fim de contornar a baixa produtividade do rebanho bovino passou-se a adotar sistemas de criação intensivos, aliado a um melhor manejo nutricional e ao uso de genótipos animais mais produtivos. Segundo a Associação Brasileira de Angus (2017) animais da raça Angus, quando criados nas mesmas condições alimentares de outras raças atingem a puberdade mais cedo, o que proporciona um ciclo produtivo mais curto gerando retorno ao investimento do produtor mais rapidamente.

A intensificação dos sistemas de produção de gado de corte no Brasil e o uso do cruzamento industrial entre taurinos (*Bos taurus taurus*) e zebuínos (*Bos taurus indicus*) tem sido empregado a fim de obter-se animais mais produtivos favorecendo a redução na idade de abate dos animais com maior peso, melhor acabamento e qualidade de carcaça, como demonstrado por Dallantonia et al. (2021). Esses autores observaram que animais cruzados F1 Angus x Nelore, tanto no sistema de pastejo quanto de confinamento, apresentaram maiores ganho médio diário (GMD) e peso vivo final (PVF) em comparação aos animais Nelore nos mesmos sistemas. Além disso, a maior deposição de gordura ocorreu durante a fase de terminação em condição de confinamento.

Além do cruzamento entre genótipos a utilização de dietas com altos níveis de concentrado durante o período de confinamento se tornou uma das estratégias para melhorar a produtividade do rebanho bovino. A ótima expressão do potencial genético para desempenho produtivo dos bovinos está diretamente ligada a ingestão de quantidades adequadas de proteína e energia. Assim a utilização de grãos na alimentação dos animais confinados é uma prática vantajosa no Brasil, já que a produção brasileira de grãos é significativamente alta (Conab, 2017).

A utilização de dietas com maiores quantidades de concentrado favorece o ganho de peso, a melhor conversão alimentar e o maior rendimento com melhor acabamento de carcaça quando comparado ao sistema extensivo de produção (Santos et al., 2016). A superioridade do genótipo cruzado sobre o genótipo Nelore foi observada por Antonelo et al. (2020) ao estudar o desempenho de animais em confinamento alimentados com dietas com 85% de concentrado com e sem acréscimo de óleo de soja. Os animais cruzados apresentaram superioridade sobre os animais Nelore no consumo de matéria seca (CMS), GMD e PVF, no entanto eles não encontraram diferença entre as dietas.

A constante busca por maior eficiência biológica dos animais, seguido de deposição mais rápida de músculo e maior cobertura de gordura tem apresentado melhoria sobre as características de carcaça e qualidade da carne (Rodrigues et al., 2019).

1.3. Qualidade da Carne bovina

A carne, um dos alimentos mais consumidos pela população mundial, apresenta alto valor biológico, sendo uma importante fonte de proteína, vitaminas, minerais e ácidos graxos (Moreira et al., 2017). Nos últimos anos os consumidores passaram a ter maior preocupação como o alimento que consomem e que é produzido, principalmente em relação ao impacto que os sistemas de produção animal causam ao meio ambiente (Moreira et al., 2017). Além disso, à medida que a renda da população aumenta surgem novos padrões de consumo, principalmente da carne bovina (Passetti et al., 2016), não só aumentando o consumo, como também a busca por produtos com maior qualidade e responsabilidade ambiental.

O conceito de qualidade de carne é extremamente amplo sendo altamente influenciado por diversos fatores da cadeia produtiva da carne. As principais características que definem a qualidade da carne são: as características sensoriais (cor, odor, sabor, textura, maciez etc.), características nutricionais (perfil lipídico, composição centesimal etc.) (Descalzo e Sancho, 2008), que podem ser afetadas de acordo com a espécie animal, raça, idade, sexo, manejo alimentar, manejo post-mortem e condições de conservação (Osório et al., 2009).

1.3.1. Características sensoriais da carne bovina

1.3.1.1. Cor

A cor da carne é a característica sensorial mais importante na decisão de compra do consumidor, já que a cor está diretamente correlacionada a “carne nova”, ou seja, o vermelho brilhante da carne é associado à uma carne fresca e de maior qualidade (Bellés et al., 2019).

A mioglobina, metaloproteína, é uma proteína composta por um grupo prostético (grupo *heme*) ligado a uma molécula de proteína globular (globina), envolvida nos processos de oxigenação do músculo, e é o principal pigmento responsável pela cor característica da carne (Lima Júnior et al., 2013), então a cor da carne será determinada pela quantidade de mioglobina ali existente. A mioglobina pode estar presente na carne na forma reduzida (desoximioglobina, de cor vermelho púrpura), oxigenada (oximioglobina, vermelho brilhante) ou oxidada (metamioglobina, marrom pardo) (Osório et al., 2009).

Segundo Boles et al. (2010) à medida que o animal envelhece a mioglobina se concentra deixando a cor da carne mais intensa. A concentração da mioglobina também difere de acordo com a espécie animal, a carne bovina, por exemplo, apresenta maior concentração de mioglobina do que as carnes suína e ovina.

A percepção de cores por cada indivíduo é muito variável, por isso além da percepção sensorial, a cor da carne pode ser avaliada instrumentalmente, seja por espectrofotômetros ou colorímetros. Para fazer essa avaliação é utilizado um espaço de cores. Um espaço de cor pode ser descrito como um método para expressar a cor usando algum tipo de notação, como por exemplo números. O espaço de cores foi desenvolvido pela *Commission Internationale l'Eclairage* (CIE) para comunicação e expressão das cores.

O espaço de cor L*a*b*, conhecido também como CIELAB é o mais popular entre os espaços de cores utilizados para avaliar as cores atualmente. O espaço de cor L* (luminosidade), a* (coordenada vermelho/verde) e b* (coordenada amarelo/azul) correlaciona consistentemente os valores de cor com a visão humana, ou seja, representa da melhor forma possível a percepção de cor pelo olho humano.

Em se tratando da carne a L* consiste na capacidade da carne de refletir a luz incidente, o a* reflete a quantidade de pigmento vermelho existente nas mioglobinas e nos citocromos C presentes na carne e o b* está associado aos carotenoides presentes na carne (Hedrick et al., 1983; Priolo et al., 2001). As coordenadas cromáticas a* e b*, que representam a mioglobina, citocromos C e carotenoides, sofrem influência bioquímica e física do processo de abate. A coordenada cromática a*, em particular é afetada pelos processos de pós abate, quando a respiração celular reduz e o oxigênio fixa-se à mioglobina, dando origem as cores mais vermelhas da carne (O'Sullivan et al., 2015). As coordenadas cromáticas a* e b* são utilizadas também para calcular o ângulo da tonalidade h° e a intensidade ou saturação da cor C*.

Monteiro et al. (2022) observaram aumento no parâmetro L* na carne de animais Nelore e cruzados ao longo do tempo de maturação da carne, sendo que os animais Nelore foram os que apresentaram os maiores valores. De acordo com Lee et al. (2008) isto ocorre pois acontecem modificações químicas transformando a desoximioglobina em oximioglobina durante o processo de oxigenação muscular. Bressan et al. (2011) ao avaliarem a carne de animais *B. indicus* e *B. taurus* e relataram valores de L* maiores para os animais *B. indicus* terminados em confinamento. Para estes autores a diferença encontrada pode estar relacionada a maior quantidade de ácidos graxos polinsaturados presentes na gordura de animais *B. indicus.*

Já para os parâmetros a* e b* os animais Nelores avaliados no estudo de Aroeira et al. (2017) foram os que apresentaram os maiores valores devido à menor estabilidade demostrada ao longo do processo de maturação por causa do acúmulo de metamioglobina. A cor é uma importante aliada na hora de se oferecer um produto de qualidade para o consumidor, por isso é tão importante que se atente para todos os fatores que podem vir a influenciá-la de forma negativa.

1.3.1.2. Maciez

A maciez é uma importante característica da carne para a aceitação do consumidor, levando-o a repetir a compra e até mesmo pagar mais pela carne de melhor qualidade (Warner et al., 2021). Resumidamente a maciez pode ser descrita como a percepção sensorial que o consumidor tem da carne, como resistência à língua, à pressão do dente na hora da mastigação, aderência e resíduo pós mastigatório, ou seja, fatores altamente subjetivos (Moreira et al., 2017).

Existem duas origens para os fatores que podem influenciar a maciez da carne, os *ante mortem* (idade, genótipo, sexo, nutrição, exercício, estresse pré-abate, presença de tecido conjuntivo, espessura e comprimento do sarcômero) e os *post mortem* (estimulação elétrica, rigor *mortis*, resfriamento da carcaça, pH, maturação, método e temperatura de cozimento) (Warner et al., 2019).

Como toda característica de qualidade, a maciez também sofre influência de alguns fatores como, por exemplo, a idade do animal. Normalmente animais mais jovens apresentam maior maciez em comparação com animais mais velhos. Os fatores que influenciam a maciez da carne são: espécie, genótipo, nutrição, manejo pré-abate, e manejo pós-abate tais como: estimulação elétrica, suspensão da carcaça, maturação da carne, condições de embalagem e até mesmo as condições de cocção.

Assim como a cor, a maciez pode ser avaliada pelo painel sensorial, e instrumentalmente, pela força de cisalhamento e índice de fragmentação miofibrilar (Adrighetto et al., 2010). A força de cisalhamento é definida como a força necessária empregada para romper as fibras da carne durante a mastigação. De acordo com a Brazil Beef Quality (2020) a carne pode ser classificada em extremamente macia, quando a força de cisalhamento for <2,0 kgf/cm^2, muito macia (<3,0 kgf/cm^2), macia (<4,0 kgf/cm^2), potencialmente macia (<5,0 kgf/cm^2), com potencial de maturação (<5,0 a 7,0 kgf/cm^2) e dura (>8,0 kgf/cm^2).

Outra característica muito importante é a suculência da carne, ou seja, a capacidade da carne em reter água durante o processo de cocção. A suculência da carne cozida é a sensação de umidade observada nos primeiros movimentos de mastigação que ocorre pela rápida liberação de líquido contido na carne. A manutenção da sensação de suculência se dá, principalmente, devido à presença gordura que estimula a salivação. A gordura intramuscular aumenta a sensação de suculência na carne, pois funciona como barreira contra perda de água durante o cozimento. A perda de água e a temperatura durante o cozimento afetam a suculência da carne (Moreira et al., 2017).

McGee et al. (2022) não observaram diferença na maciez e suculência da carne de animais cruzados (Charolês x Limousin) terminados em pastejo ou em confinamento, mesmo tendo observado que os animais terminados em confinamento apresentaram carcaças mais pesadas e com maior espessura de gordura. Já Lombardi et al. (2016) ao avaliar a qualidade da carne de animais Nelore alimentados com cana-de-açúcar picada ou silagem de cana-de-açúcar com dois níveis de concentrado (50 e 80%), relataram não haver diferença entre os níveis de concentrado para a perda por cocção (P = 0,7229). No entanto, quando se comparou as fontes de volumosos os animais alimentados com a silagem de cana-de-açúcar apresentaram 23,52% de perda por cocção e tiveram valores mais elevados para a força de cisalhamento (4,88 kgf/cm^2) em comparação com os animais alimentados com a cana-de-açúcar picada (3,61 kgf/cm^2).

1.3.2. Características nutricionais da carne bovina

1.3.2.1. Composição centesimal

A carne é um alimento que apresenta grande valor biológico e é composta principalmente de água (65 a 80 %), proteína (16 a 22 %) e gordura (3 a 13 %), mas também contêm cinzas e pequenas quantidades de substâncias nitrogenadas não proteicas (aminoácidos livres, peptídeos, nucleotídeos, creatina), carboidratos, ácido lático, minerais e vitaminas (Ordonez et al., 2005). A proteína da carne é considerada, nutricionalmente, o composto mais importante por apresentar grande disponibilidade de aminoácidos essenciais, que são importantes para o crescimento, desenvolvimento e manutenção do organismo vivo, e ainda promovem a imunidade e atuam na prevenção de doenças (Bridi, 2014). Segundo Young et al. (2013), na carne pode-se encontrar alto teor de minerais importantes à saúde humana como sódio, fósforo, potássio, ferro, zinco e magnésio. Dentre os minerais citados o ferro e o zinco, na carne, se encontram em concentrações altamente disponíveis, em comparação com outros alimentos. Já a gordura presente na carne tem função estrutural, funcional e metabólica dentro do organismo animal, além de contribuir com o isolamento térmico. Os lipídeos podem ser encontrados na carne nas formas saturada e insaturada, sendo que a forma saturada é tratada como vilã à saúde humana por levar ao desenvolvimento de doenças cardiovasculares. Todavia ainda não há relação comprovadamente clara entre o consumo de carne bovina e o aumento de doenças cardiovasculares e até mesmo de alguns tipos de câncer, como vem sendo demonstrado por estudos recentes (Lawrence, 2013; Oliveira, e Pitchel, 2013; Vahmani et al., 2015, 2020).

Segundo Ítavo et al. (2021) a composição centesimal pode sofrer alterações por causa da nutrição, idade, sexo, composição genética animal, entre outros. Entretanto apesar do sexo ter efeito sobre o ganho de peso médio diário, ganho de peso, peso ao abate e peso de carcaça, este não teve influência sobre a composição centesimal da carne. Esses autores relataram que a influência sobre a composição da carne é atribuída a idade do animal, que ainda assim é insuficiente para alterar a deposição tecidual e consequentemente a composição da carne.

O manejo nutricional tem grande importância no desenvolvimento animal. O tipo de alimento (volumoso e/ou concentrado), a qualidade e a quantidade de alimento fornecido podem favorecer o ganho de peso, peso ao abate, peso de carcaça, assim como maior deposição de gordura. No estudo de Ítavo et al. (2021) a composição centesimal da carne não sofreu alterações quando novilhos e novilhas Nelore e Beefalo receberam diferentes níveis de lipídeos na dieta. Entretanto quando a fonte lipídica fornecida está na forma protegida, as chamadas gorduras protegidas, o teor de extrato etéreo no músculo aumenta (Lima et al., 2015b), demonstrando a maior eficiência da gordura protegida em passar pelo rúmen para ser absorvida no intestino delgado e depositado no músculo (Lima et al., 2016).

Salim et al. (2019), estudaram a composição centesimal em diferentes músculos e observaram que esta foi influenciada pela fonte muscular, sendo que o músculo *Longíssimus lumborum* (LL) apresentou maior teor de proteína e menor teor de cinzas e lipídeos do que o músculo *psoas major* (PM). Em contraste, Canto et al. (2016) não observaram diferenças nos teores de proteína e cinzas nos músculos LL e PM de bovinos Nelore à pasto e relataram que o músculo LL apresentou maior umidade em comparação com o músculo PM.

Lopes et al. (2012a), ao avaliarem a composição centesimal do músculo *longíssimus thoracis* (LT) de animais Red Norte (½ Senepol x ¼ Angus x ¼ Nelore) e Nelore terminados em confinamento, não encontraram diferença (P > 0,05) entre os genótipos. Os valores encontrados para os animais Red Norte e Nelore foram, respectivamente: 74,6 e 74,2% para umidade, 1,0 e 1,0% para matéria mineral, 21,1 e 21,9% para proteína e, 2,1 e 2,3% para lipídios totais. Igualmente, Barcellos et al. (2017), que avaliaram a composição centesimal do músculo LT de animais de diferentes grupos genéticos, não observaram diferenças (P > 0,05) nos teores de umidade (72,5 e 72,4%), matéria mineral (1,2 e 1,2%), proteína (22,1 e 22,6%) e, lipídios totais (4,3 e 4,2%) entre Nelore e Nelore x Angus, respectivamente. Por sua vez, resultados semelhantes foram encontrados por Oliveira et al. (2021), ao analisarem a composição centesimal do músculo LT de animais Nelore e Nelore x Angus. Estes autores não encontraram encontradas diferenças (P > 0,05) nos teores de umidade (72,5 e 34 73,5%), matéria mineral (1,2 e 1,3%), proteína (24,6 e 24,1%) e, lipídios totais (1,7 e 1,6%) entre a carne de animais Nelore e Nelore x Angus, respectivamente.

A composição centesimal é um indicador da qualidade nutricional da carne e um indicador de como o animal está aproveitando os nutrientes fornecidos na dieta.

1.3.2.2. Perfil de ácidos graxos

O consumidor tem exigido, cada dia mais, uma carne bovina com maior qualidade. Assim, a demanda por estratégias produtivas visando atender as exigências dos consumidores tem aumentado. É desejado pelo consumidor uma carne com perfil lipídico que não traga malefícios à saúde, uma vez que a gordura é vista como precursora de problemas cardiovasculares, no entanto, não é toda gordura que causa prejuízos à saúde. Com isto, os estudos têm se voltado para a modificação do perfil de ácidos graxos da carne com o intuito de se obter menor proporção de ácidos graxos saturados, para que a carne produzida seja mais saudável e traga benefícios à saúde humana (Ladeira et al., 2014). É importante salientar que os ácidos graxos saturados atuam de forma individual e afetam a fisiologia e saúde humana de formas diferentes, e é preciso considerar que esses efeitos podem ser influenciados pelos outros componentes da dieta, como por exemplo, carboidratos e alimentos processados (Astrup et al., 2019).

Estratégias foram pensadas e testadas para obter maior qualidade dos produtos de animais ruminantes melhorando o perfil lipídico. Segundo Vahmani et al. (2020), o perfil de ácidos graxos (AG) da carne pode ser modificado pelo manejo alimentar dos animais, uma vez que o perfil de AG é altamente afetado pelos microrganismos do rúmen. Assim, a manipulação ruminal, a partir da adição de fontes lipídicas a dieta dos ruminantes, se provou funcional (Daley et al., 2010; Moloney et al., 2012). Existe uma ampla quantidade de alimentos com perfis de ácidos graxos bem diversificados que podem ser utilizados nas dietas de ruminantes como fontes de lipídeos, como por exemplo soja integral, caroço de algodão e semente de linhaça (Fiorentini et al., 2012; Oliveira et al., 2012; Ladeira et al., 2014). Todavia, existe um entrave à deposição de ácidos graxos insaturados nos tecidos de ruminantes em decorrência da biohidrogenação ruminal destes ácidos graxos (Herdmann et al., 2010). De acordo com Allen (2000), o processo de biohidrogenação pode ser potencializado em até 90% se os animais forem alimentados com dietas que tenham óleo como fonte lipídica.

Para contornar este obstáculo há a possibilidade de fornecer aos animais uma fonte lipídica conhecida como gordura protegida, que apresenta lenta liberação de ácidos graxos no rúmen (Jenkins e Bridges, 2007), o que resulta em maior quantidade de ácidos graxos insaturados chegando ao intestino delgado, resultando em maior deposição de ácidos graxos monoinsaturados nos tecidos. Sementes de oleaginosas, como os grãos de soja, quando fornecidos inteiros, também são uma fonte lipídica de liberação lenta no rúmen. A utilização de gordura protegida e grãos de oleaginosas inteiros é uma estratégia nutricional capaz de aumentar o escape ruminal de ácidos graxos insaturados e por consequência aumentar os níveis desses ácidos no tecido animal ao mesmo tempo em que há redução dos ácidos graxos saturados depositados (Duckett e Gillis, 2010).

Animais alimentados com grãos de oleaginosas, normalmente apresentam maiores concentrações de ácidos graxos monoinsaturados (AGM), destacando o ácido oleico, que é o principal AGM da carne bovina (Daley et al., 2010). Além disso, o fornecimento de grãos pode diminuir as concentrações dos ácidos graxos saturados (AGS), especialmente os ácidos láurico, mirístico e palmítico. Entretanto, o fornecimento de concentrações muito altas podem prejudicar o desempenho animal ao reduzir consumo de matéria seca (CMS) em decorrência da diminuição da digestão da fibra insolúvel em detergente neutro (FDN) da dieta.

Ladeira et al. (2014) demonstraram que ao fornecer dieta contendo soja moída, foram encontrados maiores teores de ácido linoleico (C18:2), ácido α-linolênico (C18:3) e ácido graxo poli-insaturado total no rúmen, enquanto ao fornecer uma dieta contendo gordura protegida, foram observados maiores teores dos ácidos palmítico (C16:0), esteárico (C18:0) e oleico (C18:1). Estes autores relataram ainda que as fontes de lipídios utilizadas não afetaram o conteúdo de ácido mirístico ou palmítico no músculo *Longíssimus dorsi*.

Levando em consideração a saúde humana, os resultados encontrados por esses autores apresentam importantes implicações, pois segundo Woollett, Spady, & Dietschy (1992), os ácidos mirístico e palmítico interferem, no fígado, na função normal dos receptores de LDL (*low density lipoprotein*), diminuindo a remoção do LDL e aumentando sua concentração no plasma. Os animais que foram alimentados com a soja moída apresentaram maior quantidade de CLA (cis-9, trans-11 C18:2) no músculo *Longíssimus dorsi*, e de acordo com os autores, isto deve ter ocorrido por causa de uma maior biohidrogenação do ácido linoleico em consequência da moagem da soja (Ladeira et al., 2014).

Ferrinho et al. (2018), ao avaliarem o perfil de ácidos graxos da carne de animais Nelore alimentados com dietas sem semente de algodão (C), com semente de algodão (CSA) e com semente de algodão e vitamina E (CSAE) observaram que não houve efeito da dieta sobre as concentrações dos AGS ($p>0,05$), no entanto relataram redução na concentração dos AGM da dieta C (2,340%) para as dietas CSA (1,910%) e CSAE (1,770%). Estes autores levantaram a hipótese de que essa redução na concentração do AGM pode estar associada ao fato de que a semente de algodão pode inibir a enzima Δ^9 dessaturase, que é responsável pela insaturação endógena do AGS para o AGM.

Menezes et al. (2014) avaliaram animais da raça Devon terminados em sistema de pastejo (pastagem tropical e temperada) e em confinamento, recebendo silagem de milho e concentrado a base de farelo de trigo e grão de milho. Estes autores relataram que a dieta do confinamento apresentou grande concentração de ácidos graxos insaturados (57,32%), em decorrência da presença de ácido oléico (23,28%) e linoléico (28,94%), característico de dietas com presença de grãos. A pastagem temperada apresentou alta concentração de ácidos graxos insaturados, pela presença de ácido linolênico (46,27%). Já na pastagem tropical teve alta participação de ácido linolênico (22,74%) e, de ácido palmítico (24,39%). Quando eles compararam o total de ácidos graxos insaturados (AGI) na carne dos novilhos mantidos em sistema de pastejo, foi observado que a pastagem temperada apresentou maior quantidade de AGI em relação aos animais mantidos em pastagem tropical, não havendo diferença para a carne dos novilhos terminados em confinamento (47,21;41,05 e 44,54%, respectivamente).

A adição de fontes óleo à dieta dos bovinos pode favorecer a redução dos teores de ácidos graxos saturados e o aumento dos teores de ácidos graxos insaturados, devido a composição dos próprios óleos, que apresentam maior quantidade de ácidos graxos insaturados, principalmente os ácidos graxos n-3 e n-6, como relatado por Oliveira et al. (2012). Por isso a adição de óleo a dieta dos animais proporcionou maiores teores de ácidos graxos insaturados e redução de alguns ácidos graxos saturados.

Carvalho et al. (2017) avaliaram a composição de ácidos graxos da carne e da gordura subcutânea de novilhos Nelore suplementados com óleo de palma, óleo de linhaça, gordura protegida no rúmen (óleo de soja), soja integral e sem suplementação (controle). Estes autores observaram que a suplementação com óleo de palma aumentou as concentrações de ácido láurico e ácido mirístico no músculo e na gordura subcutânea. Eles relataram que os animais suplementados com óleo de linhaça apresentaram concentrações significativamente maiores de ácido linolênico conjugado (CLA) na carne e na gordura do que os animais do tratamento controle. Os autores chegaram à conclusão de que a inclusão de óleo de palma e de ácidos graxos protegidos derivados do óleo de soja não é recomendada como método para melhorar o perfil lipídico da carne e na gordura subcutânea de bovinos Nelore.

Ao estudarem os efeitos da inclusão de fontes lipídicas na dieta de bovinos Nelore confinados, sobre características da carne, e perfil de ácidos graxos da gordura do músculo *Longíssimus dorsi*, Barducci et al. (2016), observaram que houve modificação no perfil de ácidos graxos, com diminuição dos ácidos graxos C14:1, C16:1 e C17:1 e aumento do ácido transvaccênico, C18:2, ácidos graxos poli-insaturados e da relação ácidos graxos poli-insaturados: ácidos graxos monoinsaturados. Os autores concluíram que a inclusão de lipídios na dieta, independentemente da fonte, promove melhoria na composição de ácidos graxos da carne, elevando a quantidade de ácidos graxos insaturados sem alterar as características qualitativas da carne. Carvalho et al. (2017) também relataram que a adição de fontes lipídicas na dieta não alterou os atributos físicos da carne.

Ao testar o fornecimento de quatro níveis de lipídeos pela adição de grãos de girassol, milho e soja na alimentação de novilhos e novilhas Nelore e Beefalo, Ítavo et al. (2021) observaram interação ($P<0,05$) entre os níveis de lipídeos e o sexo para o ácido mirístico (C14:0), o ácido miristoleico (C14:1), o ácido palmitoleico (C16:1ω7), o ácido linoleico (C18:2ω6), o ácido linolênico (C18:3 ω3) e o ácido graxo poli-insaturado (PUFA) na carne do músculo *Longíssimus toracicus*. À medida que os níveis de lipídeos aumentaram os ácidos linoleico (C18:2ω6), linolênico (C18:3 ω3) e PUFA também aumentaram linearmente ($P<0,05$). Estes autores encontraram maior concentração de importantes ácidos graxos para a saúde humana (linoleico e palmitoleico) nas novilhas. Isto indica que a carne das novilhas apresenta aspectos mais desejáveis em relação ao perfil de ácidos graxos em comparação a carne dos novilhos.

Assim pode-se afirmar que uma boa nutrição aliada a uma boa genética e manejo bem-feito pode melhorar o perfil lipídico da carne, acentuando a presença de ácidos graxos que trazem benefícios à saúde do consumidor. É preciso salientar que apesar de todo esforço para produzir carne de qualidade a pecuária enfrenta desafios ambientais inerentes à produção, tais como redução dos recursos naturais, desmatamentos e emissões de gases de efeito estufa.

1.4. Emissões de Gases de efeito estufa (GEE) no Brasil

O Brasil é um país classificado como em desenvolvimento apresentando uma economia complexa e dinâmica. É um país caracterizado como urbano-industrial, com o setor agropecuário em destaque na economia nacional e mundial. Quando se refere a agropecuária sustentável o Brasil é referência mundial, fazendo uso de uma abordagem integrada da paisagem, adotando práticas sustentáveis em áreas com aptidão agrícola e incentivando a regularização ambiental das propriedades rurais (Brasil, 2020).

De acordo com a Quarta Comunicação Nacional do Brasil à Convenção Quadro das Nações Unidas sobre Mudança no Clima (Brasil, 2020) em 2016 as emissões totais de GEE do Brasil, totalizaram 1.467 Tg CO_{2e} o que representou aumento de 19,4% em relação às emissões totais de 2010. Das emissões nacionais totais de GEE, o setor Agropecuária contribuiu com 33,2%, sendo as emissões brasileiras de metano (CH_4) responsável por 27,7% das emissões totais de GEE e o óxido nitroso (N_2O) foi responsável por 12,4%. Entre 2010 e 2016, as emissões totais de CH_4 e N_2O aumentaram em 3,8% e 10,7%, respectivamente.

1.4.1.Emissão de óxido nitroso (N_2O) do solo em sistema de confinamento

O N_2O é um GEE com potencial de aquecimento global 265 vezes maior do que o CO_2 (IPCC, 2020) e sua concentração na atmosfera aumenta a uma taxa de 0,73 ppb/ano (Ciais et al., 2013). Em sistemas pecuários, as emissões do óxido nitroso ocorrem, principalmente, em função da deposição das excretas animais no solo e da fertilização de pastagens com nitrogênio (N). O efetivo bovino brasileiro contribui com 33% da emissão nacional estimada de N_2O, derivadas de urina e fezes excretadas em áreas de pastagens (Brasil, 2020). Aproximadamente 80 a 95% do nitrogênio que é lançado ao solo como urina e/ou fezes de bovinos advêm do N que é ingerido (Bolan et al., 2004). O N_2O nos solos é produzido em grande parte pelo processo microbiano de desnitrificação e, em menor grau, pela nitrificação. A nitrificação é um processo aeróbico que oxida amônio (NH_4^+) em nitrato (NO_3^-), tendo o N_2O como produto intermediário não obrigatório, enquanto a desnitrificação é um processo anaeróbico que reduz NO_3^- a N_2, com formação do N_2O como um intermediário obrigatório. Com o solo em condições favoráveis à desnitrificação há maior emissão de N_2O (De Klein e Eckard, 2008).

Espera-se aumento nas emissões de N_2O nas próximas décadas devido à intensificação da pecuária brasileira que avança, concomitante, com o aumento no volume de excretas, pelo aumento no número de animais, e na utilização de fertilizantes nitrogenados, o que contribui para elevar as emissões desse gás (Smith et al., 2007).

Recente estudo demonstrou que as emissões de N_2O dependem do tipo de excreta, sendo a urina a principal fonte em ambientes temperados e tropicais (Cardoso et al., 2019). A urina em particular, apresenta maior emissão de N_2O, pois seu principal componente é a ureia, que é convertida em NH_4^+ quando depositada no solo. Já nas fezes, o N presente é essencialmente orgânico e apresenta baixa taxa de conversão a NH_4^+ (Chaviegato et al. 2015). O N presente na urina de ruminantes pode acelerar os processos de nitrificação e desnitrificação no solo e produzir N_2O, assim, diminuir a excreção urinária de N reduziria as emissões de N_2O pela urina do gado (Zhao et al., 2019). A menor emissão de N_2O encontrado para as fezes pode estar relacionado ao N presente não estar prontamente disponível para a produção de N_2O como o N da urina (ureia).

De acordo com Lessa et al. (2014), nas pastagens, as maiores emissões de N_2O acontecem durante o período chuvoso do ano, o que os leva a acreditar que a estação do ano influencia fortemente as emissões. Bastos et al. (2020) avaliaram a emissão de N_2O em pastagem nos anos de 2009, 2010 e 2013 durante estação chuvosa e relataram aumento de 0,003 kg/ha em 2009, 0,005 kg/ha em 2010 e 0,002 kg/ha em 2013, nos fluxos de N-N_2O no solo nos primeiros dias após aplicação de excretas (fezes e urina) de ovinos, mas observaram pequeno efeito além de 20 dias após aplicação. Este efeito reduzido no fluxo de N-N_2O também foi relatado por Krol et al. (2016) e Simon et al. (2018) que fizeram aplicação de excretas de bovinos em pastagens nas estações do ano na Irlanda e no Brasil, respectivamente. Estes autores constataram que as emissões de N_2O, a partir da urina, atingiram pico médio aos 10 dias após aplicação e nas fezes esse pico ocorreu de 10 a 30 dias após aplicação.

Maciel et al. (2021) avaliaram a emissão de N_2O de fezes e urina de bovinos Nelore aplicados em solo de curral de confinamento durante o período de seca e observaram baixa emissão de N_2O, apresentando os maiores picos 69 dias após a aplicação das excretas quando aconteceu três dias de eventos de chuva. Na ocasião o tratamento urina foi o que apresentou maior emissão ao ser comparado com os tratamentos fezes e controle.

As emissões de N_2O a partir dos solos são, normalmente, proporcionais à quantidade de N depositado, porém o clima, as propriedades do solo e assim como as práticas de manejo são fatores que têm relevante influência sobre as emissões de N_2O. A estação do ano é outro importante fator nas emissões de N_2O, sendo que a precipitação e a temperatura média do ar estão fortemente relacionadas às emissões de N_2O (Zanatta et al., 2014).

Segundo Alves et al. (2012) a temperatura média diária do ar é semelhante à temperatura mensurada logo após o nascer ou ao pôr do sol. Assim sendo a temperatura do ar é um fator que afeta os fluxos de N_2O observados em 24 horas. Segundo o autor é provável que nesses dois momentos o fluxo de N_2O observado represente a média diária de N_2O.

O N_2O é produzido, principalmente, a partir de processos biológicos de nitrificação e desnitrificação. Em resumo, a nitrificação é a oxidação microbiana aeróbica da amônia em nitrato (NO_3^-), enquanto a desnitrificação é a redução microbiana anaeróbica de NO_3^- a NO, N_2O e N_2. Os processos de nitrificação e desnitrificação resultam em emissões de N_2O como produto intermediário. Todavia para que estes processos aconteçam não basta apenas que a urina animal seja depositada no solo, fatores como, teor de água do solo, temperatura, teor de matéria orgânica, teor de NO_3^-, teor de amônio (NH_4^+), microrganismos, pH do solo, densidade aparente, razão C para N, N/C/P inorgânico, entre outros, interferem na ativação desses processos.

Aguilar et al. (2014) avaliaram a influência da umidade dos solos e estação do ano nas emissões de N_2O. Os resultados mostraram que os maiores fluxos N_2O foram observados nos solos mais úmidos e durante o verão e o outono. Notou-se que o fluxo N_2O cresceu à medida que a temperatura do solo aumentou, isto ocorre devido à expansão das zonas anaeróbias, que é desencadeada pela aceleração da respiração do solo, e pelo aumento da taxa de desnitrificação por unidade de volume anaeróbio (Smith et al., 2003).

Existem muitos questionamentos e, ainda são necessários estudos para compreender como as condições climáticas, características do solo e das plantas impactam nas emissões de N_2O de excretas depositadas no solo. As informações sobre emissões provenientes das excretas dos animais em solos no confinamento são ainda escassas, por isso há necessidade de se voltar o olhar para este sistema de produção, uma vez que vem aumentando a cada dia.

1.4.2. Emissão de metano (CH_4) do solo em sistema de confinamento

As fezes presentes no solo contribuem com a emissão do CH_4. A fermentação anaeróbica das fezes pelos microrganismos presentes no solo produz o CH_4 devido à presença de matéria orgânica (Mazzetto et al., 2015).

A quantidade de CH_4 emitida pelo esterco é pequena comparada com a quantidade total de CH_4 entérico produzido pelos ruminantes (Benchaar e Hassanat, 2019). No entanto, a emissão deste gás do esterco do confinamento é relevante, porque grandes volumes de esterco podem resultar em maior emissão de CH_4 (Jayasudara et al., 2016).

Segundo Choi et al. (2020) o conteúdo de carbono, umidade e temperatura são os principais fatores que influenciam as emissões de metano. A proporção de carbono/nitrogênio (CN) no esterco, é um importante fator na redução da emissão de CH_4. A alta razão CN pode promover o crescimento de populações de *archaea* metanogênicas que são capazes de atender suas necessidades proteicas e, portanto, não reagir com o teor de carbono restante do substrato, resultando em baixa produção de CH_4. Assim, reduzir a excreção de nutrientes pelos animais pode servir como uma estratégia para mitigar a emissão de metano do esterco (Mazzetto et al., 2014; Wang et al., 2014).

As fezes apresentam considerável potencial de emissão de CH_4, pois nelas contêm abundantes microrganismos metanogênicos do trato digestivo que utilizam o carbono solúvel para produção de CH_4 sob condições anaeróbicas (Hahn et al., 2018). Estudos avaliaram as emissões de CH_4 da deposição excreta sobrepostas (fezes + urina) no solo e observaram efeito insignificante ou redução nas emissões de CH_4 em comparação com as deposições de fezes e urina separadas (Liao et al., 2018; Tully et al., 2017; Zhu et al., 2021).

Lombardi et al. (2022) avaliaram a emissão de metano pelas fezes e fezes + urina depositadas em pastagens e observaram que os fluxos diários de CH_4 foram significativamente afetados pelo dia da amostragem, tipo de excreta e sua interação ($P<0,001$).

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CHAPTER 2 – ARTICLE 1

Performance and meat quality of Nellore and crossbred bulls (Angus x Nellore) finished in feedlot with two levels of concentrate in the diet

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Abstract

Context: The crossbreeding of *Bos taurus taurus* x *Bos taurus indicus* cattle has become a valuable alternative in the search for improving the performance and quality of the animals' carcass and meat. In addition, the production system adopted, and the diets provided can improve not only animal performance, but also generate a product with characteristics that are more attractive to the consumer.

Aims: The objectives of this study were to evaluate the dry matter intake (DMI), performance, and meat quality of Nellore (purebred) and crossbred (Angus vs. Nellore) fed with two levels of concentrate (65 and 85 %).

Methods: Fouty-eight young bulls of similar age (18 mo ± 2 mo) were separated in four groups based on breed and diet: Group 1 NEL 65% (n=12; initial body weight (IBW): 415.66 kg ± 21.82kg), Group 2: NEL 85% (n=11; IBW 416.23 kg ± 21.96 kg), Group 3: AN 65% (n=12; IBW 458.13 kg ± 24.59 kg), Group 4: AN 85% (n=13; IBW 463.54 kg ± 26.06 kg). They stayed on trial during 105 days (21 days of adaptation and 84 finishing days) and they were all slaughtered at the same time when we reached 84 days on feed after the adaptation period.

Key results: The crossbred animals showed greater average body weight, average daily gain, hot carcass weight, and average daily gain in carcass. There was no difference between breed or diet (P>0.05) in DMI, however the crossbred animals showed a better feed conversion.

Conclusions: Crossbred animals showed a better performance compared to Nellore. Both breeds showed a lighter meat in comparison with the literature, although the crossbred animals presented the meat lighter, to both diets, when compared to NEL fed 85% diet. Implications: However, Nellore animals’ meat was as tender and juicy as crossbred.

Abbreviations: DMI, dry matter intake; IBW, initial body weight,

Keywords: beef cattle, crossbreeding, meat quality

1. Introduction

In the global context, Brazil has the second largest cattle herd, with approximately 202 million head, and is the second largest producer, third largest consumer and largest exporter of beef (ABIEC, 2023). These figures reflect the gradual and noticeable advances that have been taking place in Brazilian livestock farming. For many years, only pasture-based production was practiced, however, there has been an increasing use of feedlot for finishing animals. The number of confined animals increased from 7.2 million in 2021 to 7.62 million in 2022, which is equivalent to approximately 18% of the total number of animals slaughtered (ABIEC, 2023).

Approximately 80% of the beef herd produced in Brazil is composed of Nellore animals (*Bos taurus indicus*), which are known for being highly adapted to tropical climate conditions. However, Brazilian livestock farming still faces challenges in improving beef production. The global demand for beef has increased, leading producers to seek ways to produce more meat in less time. At the same time as the demand for meat has increased, so has consumer demand for a higher quality product.

In order to meet market demands and consumer demands, the use of diets containing greater levels of grains has become a strategy to accelerate the production cycle and achieve product improvements. According to Santos et al. (2016), the use of diets with greater amounts of concentrate in feedlot favors weight gain, better feed conversion and greater yield with better carcass finishing, when compared to the extensive cattle production system. Antonelo et al. (2020) observed the superiority of Angus x Nellore crossbred over the Nellore when studying the performance of those animals in feedlot fed 85% of concentrate in the diet.

The use of high levels of grain in the diet is an advantageous practice in Brazil, since Brazilian grain production is significantly high (Conab, 2023). Furthermore, the use of crossbreeding between zebu breeds, which are known to be better adapted to the tropical climate and more resistant to diseases in this environment, and taurine breeds, known for their greater production potential, also helps to accelerate the production cycle and improve the quality of the meat produced, especially marbling, tenderness and juiciness, since consumers are increasingly seeking meat with these characteristics.

As observed by Dallantonia et al. (2021), Angus x Nellore crossbred animals, in the extensive and feedlot systems, had greater average daily gain (ADG) and final live weight compared to Nellore animals in the same systems. Furthermore, the crossbred animals showed greater fat deposition during the finishing phase in confinement.

With that, the objective of this research was to evaluate the intake, performance and meat quality of Nellore and Angus x Nellore crossbred young bulls, finished in a feedlot fed with two levels of concentrate in the diet. This study may provide valuable information to help improve beef production practices.

2. Materials and methods

2.1. Local, treatments and experimental design

The study was conducted at Brazilian Agriculture Research Corporation - Embrapa Maize & Sorghum (Sete Lagoas, Minas Gerais, Brazil; 19°28’S; 44°15’W, at 732 m altitude) from May through August (dry season).

Treatments consisted of two breeds Nellore (NEL) and Angus x Nellore crossbred (AN), and two diets with two concentrate levels (65 and 85 %). The animal was the experimental unit which was randomly drawn in a completely randomized design in a 2 x 2 factorial arrangement (breed x diet).

A total of 48 young bulls aged 18 months ± 2 mo were separated into four groups based on breed and diets: NEL 65% (n=12; IBW 415.66 kg ± 21.82 kg), NEL 85% (n=11; IBW 416.23 kg ± 21.96 kg), AN 65% (n=12; IBW 458.13 kg ± 24.59 kg), AN 85% (n=13; IBW 463.54 kg ± 26.06 kg). They stayed on trial during 105 days (21 days of adaptation and 84 finishing days) and they were all slaughtered at the same time when we reached 84 days on feed after the adaptation period.

2.2 Feedlot management, slaughter, and carcass traits

Early in the feedlot period, the animals were allocated in a 240 m² (20 m long x 12 m wide) feedlot with collective corrals. The pens were equipped with feeders and water troughs offered enough space to ensure animal welfare with a minimum area of 18.5 m² per animal. All animals were dewormed for endo and ectoparasites before entering the feedlot.

The animals were fed diets with different concentrate levels (65 and 85%), which were formulated using the Maximum Profit Ration 3.3 software (ESALQ, Piracicaba, Brazil). The goal was an ADG of 1.5 kg. The diet was composed of corn silage, ground corn, ground whole soybean, urea, and mineral salt (Table 1).

Table 1. Ingredients and nutrients composition of the total mixed ration (TMR) with 65 and 85 % concentrate on dry matter (DM)

Ingredients (g/kg DM)	**TMR**	
	65%	**85%**
Corn silage	350.0	150.0
Corn grain ground	530.0	620.0
Soybean	87.0	200.0
Urea	2.0	6.0
Mineral salt[1]	23.0	23.0
Nutrients (g/kg DM)		
DM[2]	460.0	634.0
Ash[3]	50.0	54.0
OM[4]	950.0	946.0
CP[5]	130.0	150.0
EE[6]	73.0	83.0
NDF[7]	310.0	240.0
ADF[8]	130.0	70.0
TDN[9]	7331.0	7731.0

[1]Amount of mineral (per kg of supplement): Phosphorous, 18 g; Calcium, 50 g; Sulfur, 10 g; Magnesium, 20 g; Sodium, 30 g; Zinc, 1303 mg; Copper, 375 mg; Iron, 500 mg; Manganese,

520 mg; Cobalt, 50 mg; Iodine, 50 mg; Selenium, 9 mg; lasalocid sodium, 450 mg.

[2]Dry matter, [3]Ash, [4]Organic matter, [5]Crude protein, [6]Ether extract, [7]Neutral detergent fiber, [8]Acid detergent fiber, [9]Total digestible nutrients (the TDN was estimated using the formula recommended by Cappelle et al. (2001), TDN (%) = 91,0246 – 0,571588 * NDF (diet).

The diet was supplied twice a day at 0800 and 1500, and the amount supplied was adjusted daily to maintain a 10% of refusals and water *ad libitum*. The animals remained in feedlot for 105 days, with a 21-day diet and facility adaptation, and 84 days for finishing. The adaptation period began with a supply of 50% roughage and 50% concentrate and gradually increased until reaching the final level of concentrate in each diet (65 and 85% concentrate based on DM).

Animal performance was evaluated monthly with body weight (BW) recorded after a 16-hour fasting (water and solid feed). ADG was calculated using Equation 1:

$$ADG = \frac{FBW - IBW}{Days\ in\ feedlot} \qquad \text{(Eq. 1)}$$

where: FBW is final body weight and IBW is initial body weight.

All animals fasted for 16 hours and were weighed a day before slaughterhouse shipment. Subsequently, the animals were refed and were shipped to a slaughterhouse the following day. Upon arrival at the beef plant, the animals were fasted from solids for 24 hours with *ad libitum* access to water and slaughtered. The carcasses were subjected to low voltage electrical stimulation (≤100V) during blood depletion and after all slaughter process they were medially divided, washed, labelled and stored in a cold chamber at 4°C, where they remained for 24 hours. All animals were slaughtered in accordance with the humanitarian slaughter procedures required by the Brazilian legislation RIISPOA (Brasil, 1997). The hot carcass weight (HCW) was immediately recorded after cleaning. Carcass yield was calculated by dividing HCW by FBW x 100 and average daily carcass gain (ADGc) was calculated using the formula: ADGc =

(HCW – (IBW x 50%)/days in feedlot).

2.3. Dry matter intake and apparent digestibility

Dry matter intake (DMI) was measured individually in eight animals from each experimental treatment (*n* = 32) and was based on the titanium dioxide (TiO_2) as an external indicator to estimate fecal production (FP). The TiO_2 was packaged in a paper cartridge and introduced directly into the animal's esophagus with the aid of a PVC applicator. Each bag was composed of 10g of TiO_2 and was administered on each animal for 12 days (Titgemeyer et al., 2001; Marcondes et al., 2006; Sampaio et al., 2011).

Fecal samples from each animal were collected daily during the last five days of indicator supply. At each collection day, the feces were oven dried at 55°C for 72 hours and then ground in a Wiley mill (Thomas Scientific, Swedesboro, NJ, USA) to pass a 1 mm screen. After griding, a composite of all samples from each animal was analyzed. The TiO_2 concentration in the stool samples was analyzed according to Sort et al. (1996). A standard curve was prepared using 0, 2, 4, 6, 8 and 10 mg of TiO_2 and the readings were performed in a spectrophotometer at a wavelength of 410 nm as described by Meyers et al. (2004). Fecal production was estimated by TiO_2 following Equation 2.

$$\mathrm{FP} = \frac{\mathrm{Supplied\ TiO2}}{\frac{\mathrm{Fecal\ TiO2}}{\mathrm{DM\ (105°C)}}} \qquad \text{(Eq. 2)}$$

where: FP = fecal production obtained by the concentration of TiO_2 (g DM/d); Supplied TiO_2 = amount of animal supplied TiO_2 (10g/d); fecal TiO_2 = amount of fecal TiO_2 (%); DM 105°C = fecal dry matter (105°C).

Fecal production and indigestible neutral detergent fiber (iNDF) were used to estimate animal dry matter intake (DMI, kg/day). The iNDF was used as an internal indicator and was obtained from the in-situ incubation of samples of feces (iNDF feces) and diet (NDFi diet) for 288h in the rumen of a cannulated animal receiving a diet composed of corn silage and concentrate (ground corn, ground whole soybean, urea and mineral core) (Detmmann et al., 2012). The DMI was calculated based on Equation 3.

$$\mathrm{DMI} = \mathrm{FP} * \frac{\text{fecal iNDF}}{\text{diet iNDF}} \qquad \text{(Eq. 3)}$$

where: DMI = dry matter intake; FP = fecal production; iNDF = indigestible neutral detergent fiber.

The dry matter digestibility (DMD) was estimated using Equation 4, and digestibility of organic matter (DOM), neutral detergent fiber (NDF) and crude protein (CP) were estimated using Equation 5 (Berchielli et al., 2006).

$$\text{Digestibility} = \frac{(\text{DMI} - \text{Excreted DM})}{\text{ingested DM}} * 100 \qquad \text{(Eq. 4)}$$

where: DMI = dry matter intake; excreted DM = excreted dry matter; ingested DM = ingested DM.

$$\mathrm{ND}(\%) = \frac{(\text{ingested} * \%\text{nutrient}) - (\text{excreted DM} * \%\text{nutrient})}{\text{ingested DM} * \%\text{nutrient}} * 100 \qquad \text{(Eq. 5)}$$

where: ND (%) = nutrient digestibility; ingested DM = ingested dry matter; % nutrient = ingested and excreted nutrient; excreted DM = excreted dry matter.

2.4. Chemical analysis

Diet, corn silage and leftovers samples were collected weekly throughout the feedlot period. These samples were dried in a forced ventilation oven at 55°C for 72 hours. After drying, the samples were ground in a 1mm Wiley mill. The bromatological analyzes of dry matter (DM), mineral matter (MM), organic matter (OM), neutral detergent fiber (NDF), acid detergent fiber (ADF), crude protein (CP) and ether extract (EE) were carried out following the food analysis methods described by Detmann et al. (2012) (Table 1).

2.5. Meat quality analysis

Meat samples were collected from the carcasses stored at a cold chamber at 4°C, between the 12^{th} and 13^{th} ribs 24 hours after slaughter. Beef steaks (~2.5 cm in thickness) were cut, vacuum packed, identified, and stored in an MA 415 Bio-chemical oxygen demand incubator (Marconi Equipamentos Ltda., Piracicaba, Brazil) for 24 hours at a constant temperature of 2°C. These samples were used for color, cooking loss and Warner-Bratzler shear force analyses.

2.5.1. Color

To perform the color analysis, the samples were removed from storage, unpacked, and identified again. Subsequently, the samples were exposed to atmospheric air for 30 minutes for myoglobin oxygenation. To determine the CIE $L^*a^*b^*$ color space, a Chroma-Meter Cr-200b colorimeter (Konica, Minolta Inc., Japan) with D65 illuminant, 10° observation standard calibrated for a white standard was used. Five readings per sample were taken to represent the entire surface of the steaks.

To determine the chroma (c^*) and hue angle (h^*) values, the L^*, a^* and b^* coordinates obtained in the colorimetric determination (MacDougall, 1994), the following equations were used (Eq. 6 and Eq. 7):

$$c* = ((a*)^2 + (b*)^2)^{0,5} \qquad \text{(Eq. 6)}$$

$$h* = \arctan(b*/a*) \qquad \text{(Eq. 7)}$$

2.5.2. Cooking loss

The same steaks used for the color analysis were used to determine the cooking loss. The steaks weighing an average 326.87 ± 55.56g was cooked in an oven at a temperature of 160-180°C until reaching an internal temperature of 71°C (Amsa, 1995). Upon reaching an internal temperature of 40°C during cooking, the steaks were flipped and kept in the oven until reaching the final cooking temperature of 71°C. The steaks' cooking temperature was monitored by a digital thermometer inserted in the geometric center of each steak. Once the steaks reached a temperature of 71°C, it were removed from the oven and left on a bench until reached room temperature and then weighed again. Results were expressed as a percentage.

2.5.3. Shear force

The shear force was obtained on the same samples that were cooked to measure the cooking loss according to Warner-Bratzler Square described by Silva et al. (2015). Eight cylindrical subsamples (1.0 x 1.0 x 2.5 mm) from each steak core were obtained from a cylindrical cut in the direction of the muscle fibers. The subsamples were then entirely sheared perpendicular to the muscle fibers with the aid of a 1.016 mm Warner-Bratzler blade (G-R manufacturing, Manhattan, USA) at a speed of 200 mm/min coupled to a TA.TXplus texture meter (Godalming, Surrey, United Kingdom). The maximum force (N) measured, and the mean value was calculated for each steak.

2.6. Statistical analysis

The statistical analysis was performed using the R software (R Core Team, 2022). Data were analyzed using the generalized linear model (Pinheiro and Bates, 2000), and based on to the statistical model:

$$Y_{ijk} = \mu + \alpha_i + \beta_j + \alpha*\beta_{ij} + \varepsilon_{ijk}$$

Y_{ijk} = It is the observation of the k-th experimental unit that received the j level of the β factor and the α level.

μ = overall mean;

α_i = It is the effect of level i of factor α (i = Nellore, Crossbred);

β_j = It is the effect of level j of factor β (j = 85 % ou 65 %);

$\alpha*\beta_{ij}$ = Effect of the interaction between α and β factors;

ε_{ijk} = Experimental error associated with repetition, where $\varepsilon_{ijk} \sim N(\mu, \sigma^2)$.

The statistical assumptions of normality and homoscedasticity were evaluated using the Shapiro-Wilk (Shapiro and Wilk, 1965) and Bartlett (Bartlett, 1937) tests, respectively. If one of the assumptions was not met, a Box-Cox transformation strategies or variance modeling was used based in the “R nlme” package (Pinheiro and Bates, 2000). In the case of data transformations, data were reverted to their natural scale (back-transformations) for presentation after analysis of variance and comparison of means.

The means of the isolated factors were compared using the F test ($P < 0.05$) when there was no significant interaction effect. Should a significant interaction occur, the Tukey test ($P < 0.05$) was used in the split comparisons.

3. Results

3.1. Animal performance

The animal performance results (Table 2), showed no significant interaction ($P>0.05$) between breed and diet for any of the evaluated parameters. However, was observed that AN breed was superior to NEL for all other variables studied, except for the variable carcass yield (CY), which NEL showed greater results.

Table 2. Effect of breed and diet (mean ± SE) on the performance of Nellore (NEL) and crossbred Angus x Nellore (AN) young bulls finished in feedlot and fed with diets containing 65 or 85% concentrate in DM

Parameter	Breed		Diet		p-Value		
	NEL (n=23)	AN (n=25)	65% (n=23)	85% (n=25)	Breed	Diet	B*D
IBW (kg)[1]	415.95±4.96	460.83±4.76	436.89±4.85	439.88±4.87	<0.001	0.655	0.726
FBW (kg)[2]	534.74±6.61	606.40±6.34	574.13±6.47	567.02±6.49	<0.001	0.449	0.752
TG (kg)[3]	118.79±3.54	149.56±3.47	141.22±3.54	127.14±3.47	<0.001	0.006	0.483
ADG (kg/dia)[4]	1.42±0.04	1.78±0.04	1.68±0.04	1.51±0.04	<0.001	0.006	0.475
ADGc (kg/dia)[5]	1.00±0.03	1.21±0.03	1.15±0.03	1.06±0.03	<0.005	0.041	0.936
HCW (kg)[6]	286.51±3.95	319.96±3.78	304.68±3.86	301.78±3.87	<0.005	0.596	0.951
CY (%)[7]	53.57±0.24	52.77±0.23	53.06±0.24	53.27±0.24	0.023	0.564	0.349

[1]IBW: initial body weight; [2]FBW: final body weight; [3]TG: total gain; [4]ADG: average daily gain; [5]ADGc: average daily gain of carcass; [6]HCW: hot carcass weight; [7]CY: carcass yield. SE: standard error.

The 65% concentrate diet had the greatest effect on total gain (TG) (P<0.05), ADG and ADGc, while the variables of IBW, FBW, HCW and CY showed no significant difference (P>0.05) between diets.

There were no interaction between breeds and diets (P>0.05) on dry matter intake (DMI kg/day), DMI as function of body weight percentage (%BW) and metabolic body weight ($MBW^{0.75}$) (Table 3). The breeds and diets showed no effect (P>0.05) on DMI (kg/day), DMI as function of body weight percentage (%BW) and metabolic body weight ($MBW^{0.75}$).

Table 3. Dry matter intake and feed efficiency (mean ± SE) of Nellore (NEL) and crossbred Angus x Nellore (AN) young bulls finished in feedlot and fed diets containing 65 or 85% of concentrate in DM

Parameter	Breed		Diet		p-Value		
	NEL (*n*=8)	AN (*n*=8)	65% (*n*=8)	85% (*n*=8)	Breed	Diet	B*D
DMI (kg/day)[1]	9.20±0.30	9.96±0.32	9.77±0.32	9.38±0.30	0.102	0.369	0.775
DMI (% BW)[2]	2.22±0.07	2.21±0.07	2.27±0.07	2.15±0.07	0.900	0.280	0.621
DMI ($g/MBW^{0,75}$)[3]	100.05±3.37	102.38±3.49	104.11±3.49	98.34±3.37	0.672	0.250	0.668
Feed efficiency[4]	0.16±0.01	0.17±0.01	0.16±0.01	0.16±0.01	0.293	0.906	0.531

[1]DMI: dry matter intake; [2]DMI (%BW): dry matter intake in percentage of body weight; [3]DMI ($g/MBW^{0,75}$): dry matter intake as a metabolic body weight; [4]Feed efficiency: Daily gain (kg)/DMI (kg); *n*: number of animals per treatment. SE: standard error.

Dry matter digestibility (DMD), organic matter (OMD), neutral detergent fiber (NDFD) and crude protein (CPD) showed no significant interaction (P>0.05) between breed and diet (Table 4). Among the breeds, there was a difference (P<0.05) for the NDFD, which the NEL showed greater digestibility than the AN animals.

Table 4. Digestibility coefficients of dry matter (DMD), organic matter (OMD), neutral detergent fiber (NDFD) and crude protein (CPD) (mean ± SE) of diets containing 65 or 85% of concentrate in DM

Parameter (g/kg)	**Breed**		**Diet**		**p-Value**		
	NEL[1]	**AN[2]**	**65%**	**85%**	**Breed**	**Diet**	**B*D**
DMD	745.4±0.75	734.5±0.75	699.2±0.75	780.6±0.75	0.611	<0.05	0.368
OMD	763.1±0.76	753.3±0.76	720.4±0.76	795.9±0.76	0.665	<0.05	0.322
NDFD	586.4±0.69	559.3±0.75	533.7±0.75	611.9±0.69	0.042	<0.05	0.777
CPD	751.2±0.97	736.5±1.01	699.5±1.01	788.2±0.97	0.411	<0.05	0.916

[1]NEL: Nellore; [2]AN: crossbred Angus x Nellore. SE: standard error.

The diets showed a significant difference (P<0.05), where the diet containing 85% of concentrate had the greatest DMD, OMD, NDFD and CPD coefficients when compared to the 65% concentrate diet.

3.2. Meat quality

In this study, there was a significant interaction (P<0.05) for brightness (L^*), intensity of red (a^*), yellow scale index (b^*), hue angle (h^*) and chroma (c^*) between breed and diet. The averages of L^* found in this study, did not differ between themselves, except for NEL breed in the 85% diet.

When the diets were compared, only the 65% diet in the AN breed showed a difference for the L^* index. No interaction was observed between breed and diet for the characteristics of cooking loss (CL) and shear force (SF). There were no significant differences among breeds nor the diets for the CL variable. As for the SF, there was a difference (P < 0.05) between the diets, with the 85% diet having the lowest values (Table 5).

Table 5. Color characteristics, cooking loss, and shear force (mean ± SE) of *Longissimus thoracis* (LT) muscle of Nellore (NEL) and Angus x Nellore crossbred (AN) young bulls finished in feedlot fed diets containing 65 or 85 % of concentrate in DM

Parameter	**NEL[1]**		**AN[2]**		**P-Value**		
	65%	**85%**	**65%**	**85%**	**Breed**	**Diet**	**B*D**
L*	46.87±1.65^{aA}	42.42±1.65^{bA}	45.56±1.65^{aB}	47.46±1.65^{aA}	0.828	0.894	0.010
a*	16.93±1.17^{aA}	13.21±1.17^{aB}	14.63±1.17^{aA}	16.44±1.17^{aA}	0.686	0.404	0.023
b*	9.34±0.99^{aA}	5.98±0.99^{bB}	7.15±0.99^{aA}	9.25±0.99^{aA}	0.584	0.523	0.011
c*	19.37±1.46^{aA}	14.59±1.46^{bB}	16.33±1.46^{aA}	18.88±1.46^{aA}	0.665	0.438	0.016
h*	28.20±1.68^{aA}	22.40±1.68^{bB}	25.65±1.68^{aA}	29.19±1.68^{aA}	0.221	0.510	0.011
CL[3] (%)	25.14±3.10	26.02±3.10	27.90±3.17	24.81±3.17	0.748	0.184	0.416
SF[4] (kg)	7.34±0.67	5.68±0.67	7.18±0.47	6.13±0.47	0.806	0.039	0.615

[1]NEL = Nellore; [2]AN = Crossbred Angus x Nellore; [3]CL = cooking loss, [4]SF = shear force. Letters A and B distinct uppercase letters in the same row, within diets, differ by Tukey’s test; and letters a and b distinct lowercase letter in the same row, within breed, differ by Tukey’s test with 5 % of probability. SE: standard error.

4. Discussion

4.1. Animal performance

It is well known that *Bos indicus x Bos taurus* crosses are superior in performance than pure *Bos indicus.* This study proved the superiority performance of AN compared to NEL animals. The *Bos indicus* x *Bos taurus* cross usually results in greater weight gain, therefore higher slaughter weight, especially when animals are fed diets with greater energy content, which can be observed in this study.

Maciel et al. (2019) evaluated NEL and AN animals and also observed that AN animals had a greater IBW (418.38 kg) in the feedlot compared to NEL animals (337.74 kg), and showed greater gain during the feedlot period (1.87 kg vs. 1.32 kg, respectively). These results occurred even though they are contemporary animals of the same age, which reinforces the advantage of crosses. The genetic difference between Angus and Nellore breeds generates high heterosis, providing higher rates of muscle tissue accumulation (Cardoso et al., 2017).

In the present study, the CY was greater ($P<0.05$) on NEL animals. It is known that the *Bos taurus* breeds have a larger gastrointestinal tract and a greater deposition of pelvic fat compared to *Bos indicus* animals (Mousquer et al., 2014), which explain a lower CY for the AN animals observed in the present study. Although the CY of NEL animals was greater than the CY of AN animals, both breeds presented CY within 50 to 55%, which is expected for non-castrated NEL and AN animals (Cruz et al., 2014).

The changes in DMI are highly correlated with the animal's body weight (Amaral et al., 2018). Even though, the AN animals were heavier than the NEL at the beginning of this experiment, they showed similar DMI (kg/day) as the NEL animals which means that AN animals had a better use of the feed once they gained more weight daily compared to NEL cattle (1.78 kg/d vs. 1.42 kg/d, respectively). Antonelo et al. (2020) reported no effect of diet on DMI, but the AN animals had greater DMI compared to NEL (9.6 kg and 8.8 kg, respectively). The DMI in the current study also was not affected by the diet on both animal breeds.

According to Carvalho et al. (2016), the maintenance requirement of taurine cattle is greater than zebu cattle, and although taurine cattle present a better performance and are more efficient, it may be related to the difference in how zebu cattle use starch. *Bos indicus* cattle are more sensitive to diets with high energy, developing metabolic problems, such as acidosis, more often than *Bos taurus* (Millen et al., 2015; Pacheco et al., 2012). Therefore, those animals present lower growth performance compared to taurine cattle.

4.2. Meat quality

Meat color characteristics are directly related to consumer choice, as it is the first characteristic observed at the time of purchase. The general consumer tends to reject meat that has a very dark color. According to Priolo et al. (2001), diet, animal age, enzymes, and even the activities carried out by the animal, are factors that may influence the meat color.

Muchenje et al. (2009) described that, in cattle, means of L^* vary from 33.2 to 41.0, means of a^* vary from 11.1 to 23.6, and the means of b^* vary from 6.1 to 11.3. However, the current study found the lowest value of L^* (42.42) was greater than the highest value (41.0) attributed to this index by the aforementioned author. On the other hand, the values of a^* and b^* varied within the ranges in the literature.

The meat color was classified by Abularach et al. (1998) into dark when $L^* \leq 29.68$, and light when $L^* \geq 38.51$. As far as red intensity, it can be considered low when $a^* \leq 14.83$, and high when $a^* \geq 29.27$. For the intensity of yellow, $b^* \leq 3.40$ when it is low, and $b^* \geq 8.28$ when it is high. It was observed that both NEL and AN meat in this study to both diets, were light meat (L^*) with low intensity red (a^*) and high intensity yellow (b^*).

Due to the interaction observed between breed and diet to the color variables L^*, a^* and b^*, meat from NEL animals fed the 85% diet showed the lowest L^*, a^* and b^* indexes, compared to meat from NEL fed 65% diet and AN animals fed both diets, which means that the meat from NEL fed 85% diet was darker in comparison to NEL fed 65% diet and AN animals fed both diets. However, the meat from NEL fed 85% diet was classified as light by Abularach et al. (1998) classification.

According to Lee et al. (2008), during muscle oxygenation modifications occur in the chemical form of deoxymyoglobin to oxymyoglobin which results in greater L^* values, which is associated to the reduction on the mitochondrial respiratory activity, providing greater oxygenation of the myoglobin molecule, which results in more oxymyoglobin formation. The results found by Monteiro et al. (2022) differ from the results found in this study. These authors reported a significative interaction ($P<0,05$) between breed and aging time to the color variables L^*, a^* and b^*, finding greater L^* values for NEL animals (46.60) compared to AN animals (42.90) and no difference between those breeds to a^* and b^* indexes.

Bressan et al. (2011) also observed greater L* values in *Bos indicus* compared to Bos taurus animals finished in confinement. The authors linked this distinction to the increased concentration of PUFA found in the intramuscular fat of *Bos indicus* animals. Andrade et al. (2010) observed a lower L^* index in meat from NEL animals compared to meat from *Red Norte* (¼ Nellore x ¼ Angus x ½ Senepol) animals (33.74 and 34.60, respectively), as observed in the present study, but the meat of the animals from their study was darker than in this current study.

The c^* and h^* indices provide a means to assess color intensity, saturation, or approximate the degree of meat browning. The discoloration of meat is indicated by a rise in c^* and h^* values, as indicated by Lee et al. (2005). The transition of color from red to yellow was elucidated using the h^* index, where greater angles signify a diminished red hue, indicating reduced color stability in the meat (Ramos and Gomide, 2007).

Considering 48 hours after slaughter, it was possible to observe that the meat from NEL 85% of concentrate had the lowest hue angle (22.40) when compared with the meat of the NEL 65% concentrate (28.20), and with the AN breed meat under the diets 65% and 85% (25.65, 29.19, respectively). Therefore, the NEL 85% animals showed the greatest color stability. Different results were found by Aroeira et al. (2017), which reported greater h* values to Nellore meat in comparison to Angus meat with one day aging.

In agreement with other studies (Monteiro et al., 2022; Teixeira et al., 2022; Sahin et al., 2021; Fiorentini et al., 2018), no difference was observed between breeds and diets as far as cooking loss (CL). Gök et al. (2019) affirms that CL may be related to the cooking method and temperature, storage time, and meat moisture. In addition, less cooking loss results in juicier meat.

The shear force is an objective measure used to measure meat tenderness, representing the force required to break the meat fibers during mastication. Meats that present a shear force above 5.9 kgf are considered not very tender (Boleman et al., 1997). The same authors classified the meat as very tender (2.3 to 3.6 kgf), moderately tender (4.1 to 5.4 kgf) and not very tender

(5.9 to 7.2 kgf). With that, NEL and AN meat from this study were classified as not very tender, regardless of diet.

Many studies have shown that *Bos indicus* meat is less tender than *Bos taurus* meat (Wheeller et al., 2010, Elzo et al., 2012, Favero et al., 2019, Monteiro et al., 2022), and as the percentage of *Bos indicus* increase in the cross, SF increases (Wright et al., 2018), which was probably the cause of AN animals in this study not showing a tender meat, compared to NEL. The action of proteolytic enzymes, calpain and calpastatin, is another factor that can affect the meat tenderness during *post-mortem*. Calpain acts degrading myofibrillar proteins, while the calpastatin function is to inhibit the calpain (Huuskonen et al., 2016). The decrease in proteolysis due to the activity of calpastatin can result in less tender meat and, consequently, higher shear force. *Bos indicus* animals present lower levels of calpain and greater levels of calpastatin, which results in decreased tenderness of the meat, as reported by Whipple et al. (1990).

5. Conclusion

The *Bos taurus* x *Bos indicus* cross showed better performance in the feedlot finishing phase than the purebred animals (NEL). The meat from NEL and AN was lighter than the averages reported in the literature, however the NEL animals fed the 85% diet showed the darkest meat color compared to NEL animals fed the 65% diet and to AN fed both diets and the AN animals were not able to produce a meat that is statistically tender and juicier than NEL animals.

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CHAPTER 3 – ARTICLE 2

Fatty acid profile of *Longissimus thoracics* muscle of Nellore and Angus x Nellore crossbred young bulls finished in feedlot with two levels of concentrate in the diet

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Abstract

Context: Factors such as diet, breed, and type of feed consumed play an important role in meat chemical composition. More specifically, ruminal manipulation through different diets is an example of how to change the fatty acid component of beef.

Aims: The objectives of this study were to evaluate the fatty acid profile of Nellore (purebred) and Angus x Nellore young bulls fed with two levels of concentrate (65 and 85 %).

Methods: Forty-eight young bulls of similar age (18 mo ± 2 mo) were separated in four groups based on breed and diet: Group 1 NEL 65% (n=12; initial live weight (IBW): 415.66 kg ± 21.82 kg), Group 2: NEL 85% (n=11; IBW 416.23 kg ± 21.96 kg), Group 3: AN 65% (n=12; IBW 458.13 kg ± 24.59kg), Group 4: AN 85% (n=13; IBW 463.54 kg ± 26.065.80 kg). They stayed on trial during 105 days (21 days of adaptation and 84 finishing days) and they were all slaughtered at the same time when we reached 84 days on feed after the adaptation period.

Key results: There was no interaction between breed or diet ($P>0.05$) in the concentration of saturated, monounsaturated, and polyunsaturated fatty acids. Although, Nellore breed showed greater amount of lauric, miristic and pentadecanoic fatty acids. Was observed significant interaction ($P<0,05$) between breed and diet in the sum of unsaturated fatty acid, where crossbred animals fed the 85% diet showed greater amount of unsaturated fatty acids.

Conclusions: Both breeds and diets showed the ability to change the meat fatty acid profile. Crossbred animals showed higher concentration of unsaturated fatty acid on LT muscle when compared to Nellore animals. The diet with 85% of concentrate provided a greater amount of unsaturated fatty acid, n-6/n-3 ratio and thrombogenicity index.

Implications: Despite the concern involving the consumption of beef because of the potential health problems, the fatty acid profile in the LT muscle showed greater amounts of fatty acids that are beneficial to human health.

Abbreviations: IBW, initial body weight,

Keywords: beef cattle, crossbreeding, fatty acid profile, human health, *Longissumus thoracics*, meat quality

1. Introduction

Beef is a protein food with high biological value, rich in minerals, fatty acids and vitamins that are important for the proper functioning of the human body. However, there is a growing concern about consuming this product, since beef consumption is often related to cardiovascular problems. The main reason for this concern is that beef contains saturated fatty acids (SFA), which are associated with increased levels of low-density lipoproteins (LDL) and increased risks of cardiovascular diseases. However, some authors (Lawrence et al., 2013; Vahmani et al., 2015, 2020) concluded in their studies that the increase in cardiovascular diseases and cancers is not related to SFA consumption.

It is known that beef is rich in SFA (30-40% of total FA), due to the biohydrogenation process carried out by ruminal microorganisms in which unsaturated fatty acids (UFA) present in food such as palmitoleic (C16:1), oleic (C18:1), linoleic (C18:2) and α-linoleic (C18:3) are converted into SFA, such as palmitic (C16:0) and stearic acid (C18:0) (Vahmani et al., 2020, Palmquist and Mattos, 2011).

Not all SFA are hypercholesteremic; stearic acid, for example, is known as neutral cholesterol because its ability to alter blood cholesterol levels is low and it is rapidly converted into oleic acid (Bridi, 2014). The biohydrogenation process does not always occur completely, because of it some fatty acids such as linoleic and α-linoleic acids, as well as some intermediate products such as vaccenic acid (C18:1 trans-11) and conjugated linoleic acid (CLA) manage to escape the rumen and reach the small intestine where they are absorbed.

The greater absorption of UFA by the intestine can lead to a better fatty acid profile in the meat, as well as favoring the increase in the concentration of CLA acid, which, according to research, has anticarcinogenic and antilipogenic characteristics (Gebauer et al., 2011, Maria et al, 2019), which is favorable to human health.

The quality and quantity of FA found in meat varies according to several factors, such as the animal sex class, type of diet, age, breed and production system (Bressan et al., 2016; Ladeira et al., 2018). In Brazil, the main breed produced is Nellore, and most of the production is done on pasture. For this reason, the hypothesis arose that the crossbreeding of zebu and taurine animals and the provision of diets with a high level of concentrate in feedlot could improve the profile of fatty acids present in beef.

With that said, the objective of this research was to evaluate the fatty acid profile of meat from Nelore and crossbred cattle (Angus x Nelore), finished in feedlot and fed two levels of concentrate in the diet.

This study may provide valuable information to help to demystify the belief that beef is not good for human health.

2. Materials and methods

2.1. Local, treatments and experimental design

The study was conducted at Brazilian Agriculture Research Corporation - Embrapa Maize & Sorghum (Sete Lagoas, Minas Gerais, Brazil; 19°28'S; 44°15'W, at 732 m altitude) from May through August (dry season).

Treatments consisted of two breeds Nellore (NEL) and Angus x Nellore crossbred (AN), and two diets with two concentrate levels (65 and 85 %). The animal was the experimental unit which was randomly drawn in a completely randomized design in a 2x2 factorial arrangement (breed x diet).

A total of 48 young bulls aged 18 months ± 2 mo were separated into four groups based on breed and diets: NEL 65% (n=12; IBW 415.66 kg ± 21.82 kg), NEL 85% (n=11; IBW 416.23

kg ± 21.96 kg), AN 65% (n=12; IBW 458.13 kg ± 24.59 kg), AN 85% (n=13; IBW 463.54 kg ± 26.06 kg). They stayed on trial during 105 days (21 days of adaptation and 84 finishing days) and they were all slaughtered at the same time when we reached 84 days on feed after the adaptation period.

2.2 Feedlot management, slaughter, and carcass traits

Early in the feedlot period, the animals were allocated in a 240 m² (20 m long x 12 m wide) feedlot with collective pens. The pens were equipped with feeders and water troughs offered enough space to ensure animal welfare with a minimum area of 18.5 m² per animal. All animals were dewormed for endo and ectoparasites before entering the feedlot.

The diets were formulated using the Maximum Profit Ration 3.3 software (ESALQ, Piracicaba, Brazil). The goal was an average daily gain (ADG) of 1.5 kg. The diet was composed of corn silage, ground corn, ground whole soybean, urea, and mineral salt (Table 1).

Table 1. Ingredients and nutrients composition and fatty acid profile of the total mixed ration (TMR) with 65 and 85 % concentrate on dry matter (DM)

Ingredients (g/kg DM)	**TMR**	
	65%	**85%**
Corn silage	350.0	150.0
Corn grain ground	530.0	620.0
Soybean	87.0	200.0
Urea	2.0	6.0
Mineral salt[1]	23.0	23.0
Nutrients (g/kg DM)		
DM[2]	460.0	634.0
Ash[3]	50.0	54.0
OM[4]	950.0	946.0
CP[5]	130.0	150.0
EE[6]	73.0	83.0
NDF[7]	310.0	240.0
ADF[8]	130.0	70.0

TDN[9]	7331.0	7731.0
Fatty acid profile (g/100g of FA)		
Miristic (C14:0)	0.16	0.11
Palmitic (C16:0)	18.26	17.35
Stearic (C18:0)	3.56	3.99
Oleic (C18:1, n-9)	29.10	28.61
Linoleic (C18:2, n-6)	41.97	43.55
α-Linolenic (C18:3, n-3)	3.61	3.10
Σ Saturated	22.24	21.63
Σ Unsaturated	76.38	77.25
Σ Monounsaturated	30.48	30.22
Σ Polyunsaturated	45.90	47.03

[1]Amount of mineral (per kg of supplement): Phosphorous, 18 g; Calcium, 50 g; Sulfur, 10 g; Magnesium, 20 g; Sodium, 30 g; Zinc, 1303 mg; Copper, 375 mg; Iron, 500 mg; Manganese, 520 mg; Cobalt, 50 mg; Iodine, 50 mg; Selenium, 9 mg; lasalocid sodium, 450 mg.

[2]Dry matter, [3]Ash, [4]Organic matter, [5]Crude protein, [6]Ether extract, [7]Neutral detergent fiber, [8]Acid detergent fiber, [9]Total digestible nutrients (the TDN was estimated using the formula recommended by Cappelle et al. (2001), TDN (%) = 91,0246 – 0,571588 * NDF (diet).

The diet was supplied twice a day at 0800 and 1500, and the amount supplied was adjusted daily to maintain a 10% refusals and water *ad libitum*. The animals remained in feedlot for 105 days, with a 21-day diet and facility adaptation, and 84 days for finishing. The adaptation period began with a supply of 50% roughage and 50% concentrate and gradually increased until reaching the final level of concentrate in each diet (65 and 85% concentrate based on DM).

2.3. Meat fatty acid profile

Muscle samples from the LT were collected between the 12th and 13th ribs, and the diet was analyzed to determine the FA profile. Meat samples were lyophilized and analyzed for the percentage of EE present. The analysis was performed according to Method 920.85 of the official Association of Analytical Chemistry (AOAC, 1990).

The FA was analyzed in a Supelco™37 standard FAME Mix (Supelco Inc., Bellefonte, PA, USA) gas chromatography. The lipids present in the LT muscle were extracted following the Folch, Lee and Sloane-Stanley (1957) methodology and esterified according to Hartman and Lago (1973). A 5mL sample of extracted lipid was concentrated in a water bath at 45°C, with nitrogen gas, proceeding with saponification with sodium hydroxide solution in 0.5 M methanol, followed by methylation with ammonium hydrochloride, methanol, and sulfuric acid. Subsequently, 5mL of hexane was added and stirred for 10 seconds to separate the esterified FA. Then, 3mL of the supernatant (hexane and methylated fatty acid) were removed and concentrated in a water bath at 45°C with nitrogen gas. At the time of injection, the extract was diluted with 1mL of hexane and 1μL of this solution was injected into a gas chromatograph with a flame ionization detector (GC 2010, Shimadzu, Kyoto, Japan), equipped with an SP 2560 (Sulpelco, Bellfonte, PA, USA) capillary column (100 m x 0.25 mm x 0.20 μm).

The chromatographic conditions were set with an oven temperature kept at 70°C for 4 minutes. Then, it was raised by 13°C per minute until it reached 175°C and maintained at this level for 27 minutes. After that, the temperature increased by 4°C per minute until it reached 215°C, where it stayed for 9 minutes. Finally, the temperature was raised by 7°C per minute until it reached 230°C and was held at this level for 5 minutes. The injector temperature was 250 °C and the detector temperature was 300 °C.

The FA were identified by comparison with retention times presented by the chromatographic standard C4:0 to C24:0. The FA concentrations were determined by the peak areas shown on the chromatogram for each acid in relation to the total area of FA and a percentage of FA was obtained using Chromquest 4.1 software (Thermo Electron, Italy).

The activity index of the enzyme Δ9 desaturase and elongase was determined based on the mathematical indices of Malau-Aduli et al. (1997), Kazala et al. (1999), and Pitchford et al. (2002), as presented below:

$$\Delta 9 \text{ dessaturase C16} = 100 \left[\frac{\text{C16:1 cis 9}}{\text{C16:1} + \text{C16:0}}\right]$$

$$\Delta 9 \text{ dessaturase C18} = 100 \left[\frac{\text{C18:1 } cis \text{ 9}}{C\text{18:1} + C\text{18:0}}\right]$$

$$\text{Elongase} = 100 \left[\frac{\text{C18:0} + \text{C18:1 cis 9}}{\text{C16:0} + \text{C16:1 cis 9} + \text{C18:0} + \text{C18:1 cis 9}}\right]$$

The atherogenicity index (AI) was calculated using the equation proposed by Ulbricth and Southgate (1991). The atherogenicity index is an indicator of cardiovascular disease.

$$\text{IA} = \left[\frac{\text{C12:0} + 4(\text{C14:0}) + \text{C16:0}}{\Sigma\text{n6} + \Sigma\text{n3} + \text{MUFA}}\right]$$

The thrombogenicity index (TI) was calculated using the equation also proposed by Ulbricth and Southgate (1991) as an indicator for the onset of coronary thrombosis.

$$\text{IT} = \left[\frac{\text{C14:0} + \text{C16:0} + \text{C18:0}}{0{,}5(\Sigma\text{MUFA}) + 0{,}5(\Sigma\text{n6}) + 3(\Sigma\text{n3}) + \left(\frac{\Sigma\text{n3}}{\Sigma\text{n6}}\right)}\right]$$

2.4. Statistical analysis

The statistical analysis was performed using the R software (R Core Team, 2022). Data were analyzed using the generalized linear model (Pinheiro and Bates, 2000), and based on to the statistical model:

$$Y_{ijk} = \mu + \alpha_i + \beta_j + \alpha{*}\beta_{ij} + \varepsilon_{ijk}$$

Y_{ijk} = It is the observation of the k-th experimental unit that received the j level of the β factor and the α level.

μ = overall mean;

α_i = It is the effect of level i of factor α (i = Nellore, Crossbred);

β_j = It is the effect of level j of factor β (j = 85 % ou 65 %);

$\alpha*\beta_{ij}$ = Effect of the interaction between α and β factors;

ε_{ijk} = Experimental error associated with repetition, where $\varepsilon_{ijk} \sim N(\mu, \sigma^2)$.

The statistical assumptions of normality and homoscedasticity were evaluated using the Shapiro-Wilk (Shapiro and Wilk, 1965) and Bartlett (Bartlett, 1937) tests, respectively. If one of the assumptions was not met, a Box-Cox transformation strategies or variance modeling was used based in the “R nlme” package (Pinheiro and Bates, 2000). In the case of data transformations, data were reverted to their natural scale (back-transformations) for presentation after analysis of variance and comparison of means.

The means of the isolated factors were compared using the F test ($P < 0.05$) when there was no significant interaction effect. Should a significant interaction occur, the Tukey test ($P < 0.05$) was used in the split comparisons.

3. Results

3.1. Meat chemical composition and quality

No interaction was observed between breed and diet for the chemical composition of the meat (Table 2). The same happened for the breed and diet, which did not show a significant effect ($P>0.05$) on meat DM, MM, CP, and EE of the NEL and AN animals.

Table 2. Chemical composition (mean ± SE) of *Longissimus thoracics* (LT) muscle of Nellore (NEL) and Angus x Nellore crossbred (AN) young bulls finished in feedlot fed diets containing 65 or 85 % concentrate in DM

Parameter	**Breed**		**Diet**		**P-Value**		
	NEL	**AN**	**65%**	**85%**	**Breed**	**Diet**	**B*D**
DM[1] (g/kg)	301.0±1.14	332.5±1.18	300.6±1.14	332.9±1.18	0.079	0.064	0.407
Ash	47.3±0.34	45.6±0.32	47.3±0.33	45.6±0.33	0.729	0.701	0.594
CP[2]	246.7±0.57	245.9±0.57	248.3±0.57	244.3±0.57	0.929	0.622	0.653
EE[3]	59.5±0.24	61.3±0.24	61.0±0.24	59.7±0.24	0.608	0.705	0.917

[1]DM = dry matter; [2]CP = crude protein; [3]EE = ether extract. SE: standard error.

3.2. Fatty acid profile

Apart from stearic FA, there was no significant interaction between breed and diet for any of the other studied FA (Table 3). There was an effect of the diet on pentadecanoic, palmitic, margaric, oleic, trans and linoleic acids, with the 85% diet having greater amounts of pentadecanoic, margaric, trans and linoleic acids, while the 65% diet showed greater amounts of palmitic and oleic acids. On the other hand, breed showed an effect on lauric, myristic, pentadecanoic, myristoleic and trans FA. Animals of the NEL breed had a greater amount of lauric, myristic, pentadecanoic and myristoleic acids, while animals of the AN breed had a greater amount of trans configuration acids.

Table 3. Composition of the primary fatty acid (mean ± SE) present in *Longissimus thoracis* (LT) muscle of Nellore (NEL) and Angus x Nellore crossbred (AN) young bulls finished in feedlot fed diets containing 65 or 85 % concentrate in DM

Parameter		**Breed**		**Diet**		**P-Value**		
	Carbon	**NEL**	**AN**	**65%**	**85%**	**Breed**	**Diet**	**G*D**
Saturated fatty acids								
Lauric	C12:0	0.09±0.004	0.07±0.004	0.07±0.004	0.08±0.004	<0.001	0,582	0,875
Myristic	C14:0	2.88±0.09	2.45±0.09	2.67±0.09	2.66±0.09	<0.001	0.919	0.404
Pentadecanoic	C15:0	0.43±0.02	0.37±0.02	0.37±0.02	0.42±0.02	0.031	0.036	0.913
Palmitic	C16:0	22.93±0.23	22.57±0.23	23.28±0.23	22.22±0.23	0.282	0.003	0.724
Margaric	C17:0	0.84±0.03	0.83±0.03	0.76±0.03	0.90±0.03	0.879	0.002	0.435
Stearic	C18:0	12.31±0.61	12.65±0.61	12.25±0.61	12.71±0.61	0.585	0.459	0.045
Monounsaturated fatty acids								
Myristoleic	C14:1 (n-5)	0.86±0.05	0.69±0.05	0.85±0.05	0.72±0.05	0.019	0.067	0.851
Palmitoleic	C16:1 (n-7)	3.43±0.21	3.19±0.21	3.51±0.26	3.12±0.12	0.737	0.180	0.387
Heptadecenoic	C17:1 (n-7)	0.69±0.03	0.67±0.03	0.67±0.03	0.69±0.03	0.626	0.733	0.162
Oleic	C18:1 (n-9)	35.55±0.85	36.64±0.81	37.52±0.78	34.59±0.88	0.358	0.019	0.880
Cis-Octadecenoic[1]	C18:1	4.91±0.14	4.93±0.14	5.01±0.14	4.83±0.14	0.929	0.380	0.622
Trans-Octadecenoic[2]	C18:1	3.13±0.31	3.93±0.31	2.42±0.13	4.63±0.42	0.018	<0.001	0.606
Polyunsaturated fatty acids								
Linoleic	C18:2 (n-6)	6.76±0.66	6.33±0.62	5.49±0.55	7.74±0.73	0.635	0.019	0.386
CLA – Rumenic acid	C18:2 (*cis*-9, *trans*-11) (n-7)	0.37±0.03	0.43±0.03	0.37±0.03	0.42±0.03	0.229	0.247	0.762
α-Linolenic	C18:3 (n-3)	0.45±0.04	0.48±0.03	0.44±0.03	0.49±0.03	0.162	0.207	0.698
Dihomo- γ-Linoleic	C20:3 (n-6)	0.16±0.02	0.13±0.02	0.15±0.02	0.15±0.02	0.265	0.931	0.553
Arachidonic	C20:4 (n-6)	1.01±0.14	0.88±0.12	0.89±0.12	0.96±0.14	0.429	0.693	0.657
Eicosapentaenoic	C20:5 (n-3)	0.19±0.03	0.14±0.02	0.16±0.03	0.17±0.03	0.203	0.776	0.615

Docosapentaenoic	C22:5 (n-3)	0.17±0.06	0.08±0.04	0.15±0.05	0.10±0.04	0.196	0.498	0.377
Docosahexaenoic	C22:6 (n-3)	0.05±0.01	0.03±0.01	0.04±0.01	0.04±0.01	0.085	0.787	0.817

[1]Sum of C18:1 cis 11, 12, 13, 15; [2]Sum of C18:1 trans 6, 7, 8, 9, 10, 11, 12. SE: standard error.

The FA that showed the greatest concentration in both breeds were palmitic, stearic, and oleic. They represented 71.88% and 69.60% of all FA present in the meat of NEL animals fed with 65% and 85% diets, respectively. Moreover, these FA represented 74.22% and 69.42% of all fatty acids present in the meat of AN animals fed diets 65 and 85%, respectively.

The sum of UFA and the thrombogenicity index (TI) were the only evaluated parameters to show significant interaction ($P < 0.05$) between breed and diet (Table 4). The NEL animals consuming the 85% diet differed from NEL animals consuming the 65% diet and AN animals consuming both diets. The AN animals consuming the 65% diet had the lowest percentage of UFA compared to the NEL animals consuming the 85% diet. In IT, the NEL animals consuming the 65% diet were the ones that presented the lowest index when compared to the NEL animals consuming the 85% diet and the AN animals consuming both diets.

Table 4. Sum and fatty acid proportion, enzymatic activities, and atherogenicity and thrombogenicity index (mean ± SE) present in *Longissimus thoracis* muscle of Nellore (NEL) and Angus x Nellore crossbred (AN) young bulls finished in feedlot fed diets containing 65 or 85 % concentrate in DM

Parameter	NEL		AN		P-Value		
	65%	85%	65%	85%	Breed	Diet	B*D
Σ SFA[1] (%)	39.00±0.68	39.82±0.69	39.75±0.69	31.10±0.67	0.479	0.539	0.081
Σ UFA[2] (%)	59.51±0.57[aA]	58.66±0.57[bA]	58.08±0.54[aB]	60.30±0.54[aA]	0.851	0.168	0.011
Σ MUFA[3] (%)	49.38±1.19	48.62±1.28	51.65±1.28	49.36±1.19	0.267	0.230	0.541
Σ PUFA[4] (%)	1.55±0.04	1.58±0.04	1.65±0.04	1.60±0.04	0.672	0.099	0.358
Σ n-6 (%)	7.26±1.07	8.51±1.22	5.91±0.91	9.04±1.27	0.645	0.057	0.378
Σ n-3 (%)	1.28±0.20	1.10±0.18	0.98±0.18	1.10±0.19	0.479	0.835	0.413
n-6/n-3	5.83±0.23	9.42±2.38	6.11±0.25	8.31±0.42	0.636	<0.001	0.484
PUFA/SFA[5]	0.15±0.02	0.17±0.02	0.13±0.02	0.18±0.02	0.606	0.116	0.484
Δ^9 desaturase 14	24.90±1.25	21.22±1.25	22.55±1.25	21.10±1.25	0.331	0.050	0.385
Δ^9 desaturase 16	13.74±0.35	12.10±0.67	11.12±2.50	12.33±0.45	0.050	0.065	0.313
Δ^9 desaturase 18	76.24±1.36	71.85±1.36	74.20±1.36	73.88±1.36	0.301	0.876	0.145
Elongase	66.34±0.61	66.83±0.61	68.13±0.61	67.38±0.61	0.527	0.398	0.320

AI[6]	0.59±0.02	0.59±0.02	0.57±0.02	0.53±0.01	0.006	0.046	0.165
TI[7]	1.27±0.03^{bB}	1.36±0.03^{aA}	1.36±0.03^{aA}	1.31±0.03^{aA}	0.182	0.175	0.017

^{a,b} Distinct lowercase letters in the same row, within breed, differ by Tukey's test; ^{A,B} Distinct uppercase letters in the same row, within diets, differ by Tukey's test at 5 % of probability; [1]SFA = saturated fatty acid; [2]UFA = unsaturated fatty acid; [3]MUFA = monounsaturated fatty acid; [4]PUFA = polyunsaturated fatty acid; [5]MUFA/SFA = polyunsaturated/saturated fatty acid ratio; [6]AI = atherogenicity index; [7]TI = thrombogenicity index. SE: standard error.

There was no significant (P>0.05) difference between breeds and diets for the sum of SFA, monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA). The proportion of omega 3 (n-3) did not differ (P>0.05) between breeds and/or between diets. There was a significant (P<0.05) difference between the diets for the proportion of omega 6 (n-6), in which the 65% diet had the lowest concentrations (7.26 and 5.91) for NEL and AN, respectively, compared to the 85% diet (8.51 and 9.04). The studied diets showed a difference (P<0.05) for the n-6/n-3 ratio, which the 65% diet presented the lowest values, both for NEL (5.83) and for the AN animals (6.11), in comparison with the 85% diet, which presented values of 9.42 for the NEL breed and 8.31 for the AN breed. No difference (P>0.05) was observed between breeds and/or diets for the PUFA/AGS ratio.

There was no interaction (P > 0.05) between breed and diet regarding the activities of Δ9 desaturase enzymes 14, 16 and 18. Equally, no differences (P>0.05) were found between breeds for Δ9 desaturase 14 and 18 enzymes and no differences were found between diets for Δ9 desaturase 16 and 18 enzymes. The Δ9 desaturase 14 enzyme was significantly different (P = 0.05) between diets, and the 65% diet showed the greatest activity value. The enzyme Δ9 desaturase 16 demonstrated a significant difference (P = 0.05) among the breeds, with a greater activity observed in NEL animals (12.90) compared to AN animals (11.72).

There was no interaction between breed and diet regarding elongase enzyme (P > 0.05), and no significant differences were observed between breeds and diets for this enzyme. No interaction was observed between breed and diet for AI. Nonetheless, there was a significant difference ($P<0.05$) between the breeds for AI index, and the meat of AN animals, fed with the 85% diet had the smallest AI (0.53) compared to NEL animals fed the 85% diet. The TI showed an interaction ($P < 0.05$) between the breed and the diet, and the meat of the NEL 65% diet presented the smallest TI (1.27) compared to NEL 85% diet and AN animals fed with the 65 and 85% diets.

4. Discussion

4.1. Meat Chemical composition

The meat chemical composition and quality was not affected by treatments. Macedo et al. (2022) used a centesimal scale and showed that fat is the component that suffers the greatest variation. The same research concluded that the amount of deposited fat is normally related to the balance between energy and the required dietary metabolism. However, the authors found no difference in fat content between the studied breeds.

Lopes et al. (2012) analyzed the chemical composition of the *Longissimus dorsi* muscle of Nellore and *Red Norte* (½ Senepol x ¼ Nellore x ¼ Angus) animals finished in feedlot and found no difference ($P>0.05$) between breeds for moisture content (74.6% and 74.2%), mineral matter (1.0% and 1.0%), protein (21.1% and 21.9%) and ether extract (2.1% and 2.3%). The authors associate the lack of difference in the meat chemical composition to the homogeneity of the production system, having animals of the same sex, consuming the same diet, and finished in the same system. In the present study, the production conditions were also homogeneous with animals of the same sex, contemporaneous and finished in the same system. Although the supplied diet was not the same for all animals, the roughage:concentrate ratio did not cause changes in the chemical composition of the meat.

4.2. Fatty acid profile

The concentration of oleic, palmitic, and stearic fatty acids is found greater in beef, compared to other meat. With that being said, a growing concern is arising on overconsumption of diets that are rich in SFA and poor in UFA, as the excess can bring problems to human health such as obesity, cancer, and cardiovascular diseases. Most SFA are considered hypercholesteremic, with lauric, myristic and palmitic acids being the most relevant (Briggs et al., 2017). In the present study, the meat of NEL animals had a greater percentage of lauric acid compared to the meat of AN. Micha and Mozaffarian (2009) demonstrated that lauric acid could lead to an elevation in low-density lipoprotein (LDL) levels and impact high-density lipoprotein (HDL), resulting in a decrease in the total cholesterol to HDL ratio. However, the levels of lauric acid observed in this study were relatively low for both breeds, a finding deemed normal for beef cattle according to Liu et al. (2020).

Each SFAhas a distinct effect on the concentration of cholesterol fractions of plasma lipoproteins (FAO/WHO, 2010). For example, the lauric (C12:0), myristic (C14:0) and palmitic (C16:0) acids increase LDL, while stearic acid (C18:0) has no effect. However, Liu et al. (2020) reported that stearic acid compared to the other SFA, reduced plasma LDL levels and did not affect HDL, which opposes FAO/WHO (2010). Monteiro et al. (2022) also observed greater levels of lauric and myristic acids levels for NEL animals (0.09) when compared to AN animal (0.05). Lopes et al. (2012) observed a greater concentration of lauric acid in the meat of NEL, compared to Red Norte animals (0.08 and 0.07, respectively).

Among the SFA, stearic acid highlights as the one in greater quantity. This FA is considered neutral for human health and can reduce LDL due to its ability to elongate the molecular chain, transforming stearic acid into oleic acid (Araújo et al., 2021). Thus, explaining the concentration of stearic acid in this study being lower than oleic acid in the meat. Monteiro et al. (2022) also reported a lower concentration of stearic acid and greater concentration of oleic acid when evaluating meat from NEL and AN animals. The authors associated their findings to the greater activity of the Δ9 desaturase enzyme.

The studies from the mid-20th century recommended reducing the consumption of SFA because of the risk of increased serum total cholesterol and LDL (Vahmani et al., 2020). However, new studies have questioned these recommendations, as the intake SFA is not necessarily related to an increased risk of cardiovascular disease (Renna et al., 2019; Visioli and Poli, 2020, Vahmani et al., 2020). With that, to comprehensively grasp the potential for modifying the blood profile of FAs, it is essential to examine them on an individual basis.

The CLA is exclusively present in animal products, originating from the incomplete biohydrogenation of dietary FAs and the endogenous synthesis of vaccenic acid through the action of the enzyme Δ9 desaturase in adipose tissue (McAffe et al., 2010). It is estimated that over 80% of the CLA found in ruminant fat is produced endogenously through the activity of the Δ9 desaturase enzyme (Palmquist et al., 2005).

Several studies have been carried out in order to evaluate the benefits of CLA. The CLA presented properties capable of acting in the reduction of body fat, blood triglycerides, cancer, cardiovascular diseases, prevention of fatty liver, in addition to modifying the immune and inflammatory response of the organism (Gebauer et al., 2015; Vahmani et al., 2020). However, no difference was observed in the concentration of CLA between the breeds and diets tested in this study.

The greater UFA concentration in both NEL and AN meat's may be related to the greater proportion of concentrate present in the diet; since high concentrate diets reduce the ruminal pH, which reduces the biohydrogenation of FA. Therefore, aid the UFA absorption in the post-rumen. Feeding high concentrate diets in feedlots increases the rate of passage through the rumen, which may increase UFA escape from the rumen (Jenkins et al., 2008).

The lower concentration of SFA is linked to the greater ability of these acids to be desaturated by the action of Δ9 desaturase enzymes. The greater concentration of UFA in animal meat is due to desaturation of SFA for UFA synthesis. According to WHO (2003) there is evidence that the replacement of SFA by PUFA in the diet decreases the risk of coronary heart disease.

It is known that the regular consumption of n-3 is beneficial to human health as it helps to prevent diseases such as cancer, arthritis and even depression. When n-3 is incorporated into body tissue cells, it acts in cell signaling and gene expression processes (Surette, 2008). There was a significant difference between diets for the proportion of omega 6 (n-6). The 65% diet had the lowest concentrations with omega 6 levels of 7.26 and 5.91, compared to the 85% diet with omega 6 levels of 8.51 and 9.04 for Nellore and crossbreed, respectively. The FA n-3 and n-6 are necessary because they are responsible for the maintenance of cell membranes, brain functions and transmission of nerve impulses, in addition to participating in the synthesis of hemoglobin and cell division (Martin et al., 2008).

The n-6/n-3 and PUFA/SFA ratios are used to assess the nutritional value of fat for human consumption. The great n-6/n-3 ratio found in this study may be linked to the diet supplied to the animals. In this case, containing a high percentage of concentrate since a greater grain intake result in a lower concentration of n-3 in the meat (Calabro et al., 2014), which occurred in this study. According to FAO/WHO (2010) there is no specific consumption recommendation for the n-6/n-3 ratio, if the consumption of n-3 and n-6 fatty acids are within the recommended range. However, the PUFA consumption should range within 6 and 11%, with total n-3 intake between 0.5 and 2% and n-6 between 2.5 and 9%. It is observed that the proportions of n-3 and n-6 found in the meat of NEL and crossbred animals are within the recommended range for consumption (FAO/WHO, 2010). Thus, beef consumption would supply the recommended daily intake in human diets.

Red meat is the single ingredient among several others in the human diet. Thus, one should consider the triglyceride content of the total diet consumed aside from just the meat. The recommended approach for consuming these FAs is to consider the amount of n-3 present in food and subsequently focus on the ratio of n-6 to n-3. Furthermore, to prevent diseases, a 20 to 30% daily consumption of total fat is recommended, and the UFA be around 10%.

Due to the hydrogenating action of ruminal microorganisms, meat from ruminant animals has a low PUFA/SFA ratio. In the present study, values of 0.15 and 0.17 were found for NEL animals fed diets containing 65 and 85% concentrate, respectively, and 0.13 and 0.18 for AN animals fed diets containing 65 and 85 % of concentrate, respectively, which were not statistically different. In agreement with the values observed in this study, Monteiro et al. (2022) observed a low PUFA/SFA ratio, finding values of 0.13 for AN animals and 0.17 for NEL animals. It is recommended that the PUFA/AGS ratio be between 4 to 5 and the consumption of trans fatty acids should not exceed 1% (FAO/WHO UN, 2010).

Due to the great activity of Δ9 desaturase 16, the 85% diet had the lowest proportion of palmitic acid in the meat (22:22). The enzyme Δ9 desaturase 16 acts on the desaturation of the palmitic acid chain to form palmitoleic acid. In lipogenesis, palmitic acid is a product of lipid synthesis and is rapidly converted to palmitoleic acid by the action of the enzyme Δ9 desaturase 16 or is incorporated into stearic acid by the action of the enzyme elongase. The stearic acid formed by chain elongation of palmitic acid can undergo the action of the enzyme Δ9 desaturase 18 to form oleic acid. Although there was no difference between the diets regarding the activity of the Δ9 desaturase 16 and Δ9 desaturase 18 enzymes, a considerable difference between the diets was demonstrated in the concentration of oleic acid. Bressan et al. (2016) reported greater Δ9 desaturase 16 enzyme activity in the intramuscular fat of *Bos indicus* (10.30%) than *Bos indicus* x *Bos taurus* crossbred animals (9.52%). Gama et al. (2013) also reported that *Bos indicus* has a stronger influence on its crossbreed offspring than *Bos taurus*, suggesting a genetic dominance effect.

Lopes et al. (2012) observed greater activity of the elongase enzyme by NEL compared to *Red Norte* animals, which differs from the current study. The same authors reported that the greater activity of the enzyme elongase in NEL animals can be explained by the lower values of palmitic and palmitoleic acids, and the greater content of oleic acid found in the meat of these animals. Therefore, supporting that NEL animals are carrying out greater biosynthesis of FA, mainly oleic.

The atherogenicity (AI) and thrombogenicity (TI) indexes indicate the risk of cardiovascular disease and coronary thrombosis, respectively. Encouraging beef consumption is advisable due to the presence of FAs beneficial to human health, such as oleic acid and CLA. The latter is exclusively found in animal products, primarily in ruminants. Moreover, many consumers lack essential information about the nutritional value of beef. While the consumption of meat, especially red meat, is often recommended to mitigate the risks of obesity, cancer, and metabolic syndromes, this valuable nutritional aspect is not widely recognized among the public (Biesalski, 2005).

5. Conclusion

The chemical composition of the meat was not affected by the genetic groups or the consumed diets. In this study, it was possible to observe that the breed and diet favored the change in the fatty acid profile of the meat. Both breeds fed with diets 65% and 85% had meat with a considerable profile of fatty acids in the *Longissimus thoracics*.

A hypothesis was raised that the crossbred animals had a better fatty acid profile compared to the Nellore animals; however, this possibility was only true for the concentration of UFA in the LT of the crossbred animals. The hypothesis that a diet containing 85% concentrate would provide a better fatty acid profile showed that this diet provided a greater amount of UFA, a greater n-6/n-3 ratio, and a greater thrombogenicity index.

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CAPÍTULO 4 – ARTIGO 3

Methane and nitrous oxide emissions from excreta of different beef cattle breed fed different nutritional level and finished in feedlot under tropical conditions

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Abstract: This study evaluated methane (CH_4) and nitrous oxide (N_2O) emissions from beef cattle excreta from Nellore or crossbred fed 65 or 85% of concentrated feed. The excreta were applied to feedlot floor pens and N_2O and CH_4 fluxes were monitored for 84 days in static chambers. The animals were confined for 106 days where the animal intake and performance were determined. In addition to average fluxes, the emission ratios per kg DM intake (E/DMI) and per kg weight gain (E/ADG) were estimated. The data were subjected to variance analysis in 2 x 2 factorial arrangement and means were compared by Tukey test ($P<0.05$). CH_4 emissions were 2.4 times higher in crossbred compared to Nellore animals ($P<0.039$). N_2O emissions from excreta were 50.1% lower in animals consuming diet with low compared to high nutritional level. The E/DMI and E/ADG of CH_4 were higher in crossbred compared to Nellore animals. The E/ADG and E/DMI of N_2O of animals fed high nutritional level were also 18.2% and 36.1% higher in crossbred compared to Nellore animals. The E/DMI and E/ADG of N_2O of crossbred and Nellore were also lower when fed low compared to high nutritional levels. The results showed that crossbred animals consuming diets with more concentrated feed emit more CH_4 and N_2O in their excreta. These data indicate that the Nellore herd, which is predominant in Brazil, emits lower GHGs than other breeds and that diets with high concentrate feed inclusion have greater potential to emit GHGs.

Keywords: Angus, global warming, greenhouse gases, Nellore, livestock

Introduction

The increase in greenhouse gas (GHG) emissions has already warmed the planet temperature by 1.5°C since the pre-industrial era, which has become a major concern for the world population (Intergovernmental Panel on Climate Change - IPCC, 2018). This warming could have major negative impacts on the planet due to changes in agricultural production, human survival, city infrastructure and ecosystem biodiversity (IPCC, 2023). The main GHGs are carbon dioxide (CO_2), methane (CH_4) and nitrous oxide (N_2O), with agricultural production, forestry and other land use activities contributing for approximately 23% (12.0 ± 2.9 $GtCO_2$ eq./year) of total net anthropogenic emissions between 2007 and 2016 (IPCC, 2019).

GHG emissions from cattle excreta can contribute significantly to these emissions linked to land use. Beauchemin et al. (2010) and Beauchemin et al. (2011) carried out life cycle assessment studies and observed that CH_4 and N_2O emissions from confined beef cattle excreta in Canada represented 5.00 and 22.9% of the system's total emissions. In cattle reared on pasture in Brazil, Cerri et al. (2016) observed that CH_4 and N_2O emissions from excreta represented approximately 1.40 and 25.0% of the system's total emissions. These data show the significant contribution of excreta as emissions sources to the system's total emissions, but little is known about the factors that influence this emissions type.

The CH_4 emission from bovine excreta normally varies between positive and negative fluxes (Maciel et al., 2021; Teixeira et al., 2023). These fluxes are determined by methanogenesis and methanotrophy processes (Nichols et al., 2016). Methanogenesis can occur soon after excreta application due to fermentation still taking place from the gastrointestinal tract (Angel et al., 2012; Mazzetto et al., 2014) or by soil bacteria that grow in anaerobic environment. These bacteria have increased growth due to the high nutrients supply in excreta, neutral pH and high temperatures (IPCC, 2006). Thus, variations in nutrient concentrations in excreta, which can be influenced by animal's digestion efficiency and nutrients concentration in consumed diet, can influence CH_4 emissions from excreta. To the best of our knowledge, there are no studies that have evaluated the effect of different diets and breeds on CH_4 fluxes in the excreta of beef cattle confined in tropical conditions. Teixeira et al. (2023), for example, did not observe difference in N_2O emission fluxes between Nellore and crossbred animals (Nellore x Angus), but they did not evaluate CH_4 emissions.

The denitrification and nitrification processes are mainly responsible for soil N_2O formation. The main process is denitrification, a microbial process regulated by temperature, oxygen concentration and the presence of soil organic matter labile fractions and nitrogen (Baggs and Phillipot, 2010; Smith, 2017). Climatic factors such as the occurrence of rain and high temperatures, local factors such as compaction of feedlot floor pens and animal factors such as excreta nutrients concentration can influence N_2O emissions. Teixeira et al. (2023) observed no difference in N_2O fluxes between excreta from Nellore and crossbred animals (Nellore x Angus), but emissions per kg of weight gain were higher in Nellore animals. Although the study of Teixeira et al. (2023) was the first study to evaluate breed effect, nutrient concentrations in excreta were not evaluated. Furthermore, no studies have yet been carried out to assess how nutrient concentrations in diets can affect N_2O emissions.

The rain occurrence is one of the main factors that generate peaks in N_2O emissions from excreta due to soil anaerobiosis generation (Teixeira et al., 2023). Thus, as beef cattle feedlots in tropical regions are often developed in dry season, emissions are probably lower than those estimated by the IPCC for emissions inventories purposes. This possible difference justifies further studies to determine N_2O and CH_4 emissions from excreta from beef cattle confined in tropical regions. Furthermore, there is little actually measured information on the effects of diets and breeds used in beef cattle feedlot in tropical climates. Due to the better crossbred animals' efficiency (Maciel et al., 2019; Antonelo et al., 2020) in relation to Nellore and the higher concentration of nutrients in diets recently used in commercial feedlots (Silvestre and Millen, 2021), it is possible that these factors alter substrate concentrations for N_2O and CH_4 emissions. This study aimed to determine CH_4 and N_2O emissions from the excreta of beef cattle of different breeds and fed different nutritional levels finished in feedlots in tropical conditions.

Material and Methods

All evaluations were approved by the ethics committee on animal use of Universidade Federal de Minas Gerais, under protocol number 71/2019.

Experimental site characterization

The experiment was carried out at Embrapa Maize and Sorghum, located in the municipality of Sete Lagoas, in the state of Minas Gerais, Brazil (19°28’S, 44°15’W, at 732 m altitude). According to Köppen-Geiger, the region climate is classified as Cwa, humid subtropical, with dry winters and hot and rainy summers (Alvares et al., 2013). During the experimental year, the average monthly precipitation was 143mm and the average air temperature was 21.8ºC (Figure 1).

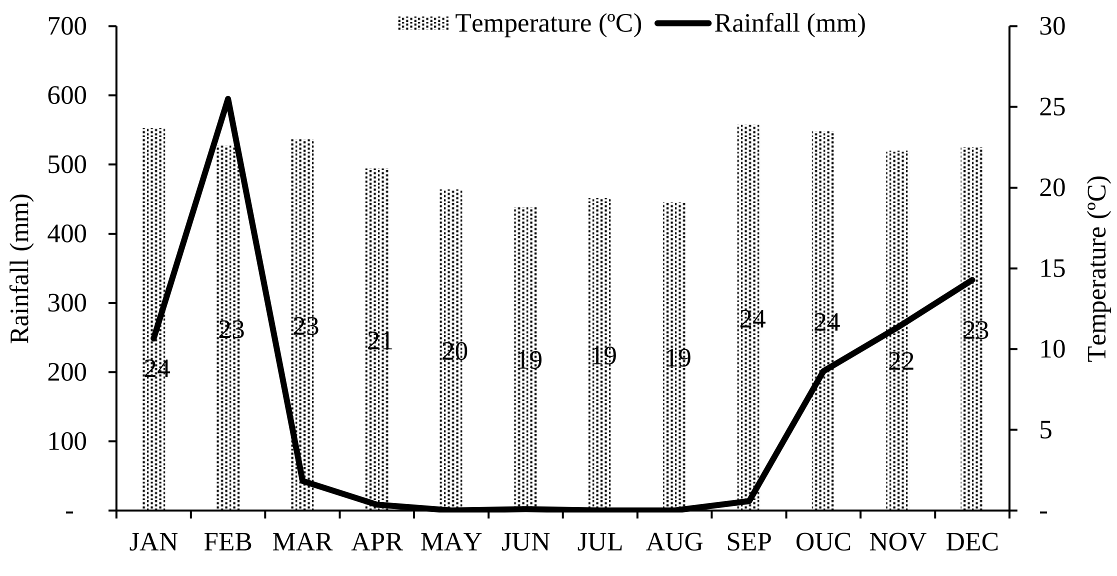

Figure 1: Average monthly precipitation and average air temperature in Sete Lagoas – MG during the experimental year

Experimental design and diets

The animals were received at 8 months of age and reared until the beginning of confinement at 18 months on *Urochloa brizantha* cv. Piatã rotational pasture under the same climatic, sanitary and nutritional conditions. The total feedlot period lasted 106 days, with the first 21 days being used for cattle adaptation. The treatments were two breeds (Nellore or crossbred - Nellore x Angus) and two diet concentrate inclusion levels (65 or 85%). A total of 40 bulls [20 Nellore, initial body weight (IBW) = 377kg ± 4.37kg and 20 crossbred breed male cattle, IBW = 415kg ± 4.20kg] were divided into four groups: 10 Nellore fed 65% concentrated feed, 10 Nellore fed 85% concentrated feed, 10 crossbred fed 65% concentrated feed and 10 crossbred fed 85% concentrated feed. Each group was distributed in pens with 18.5 m^2 per animal, with free access to diet and water. The starter diet had 50:50 roughage to concentrate ratio on dry matter (DM) basis, being increased to 25:75 ratio over three weeks. The cattle were fed three times per day (0800, 1100 and 1500 h) and adjusted daily to maintain 5.00 to 10.0% refusals. The diets composition and used feeds are described in table 1. Diet samples were collected monthly and used to determine the contents of DM, ash, crude protein (CP) and ether extract (AOAC, 1995). Neutral detergent fiber, acid detergent fiber and acid detergent lignin were determined according to Van Soest et al. (1991).

Table 1. Proportion of ingredients used (g/kg DM) and chemical composition (g/kg DM) of the diets used containing 65 or 85% concentrate in DM

Feeds	Nutritional level		Chemical composition[2]	Nutritional level	
	65%	85%		65%	85%
Whole corn plant silage	350	150	DM	460	634
Ground corn	538	621	Ash	50.0	54.0
Soybean meal	87.0	200	OM	950	946
Urea	2.00	6.00	CP	130	150
Mineral nucleus[1]	23.0	23.0	EE	73.0	83.0
			NDF	310	240
			ADF	130	70.0
			TDN	733	773

[1]Phosphorus, 18 g; Calcium, 50 g; Sulfur, 10 g; Magnesium, 20 g; Sodium, 30 g; Zinc, 1303 mg; Copper, 375 mg; Iron, 500 mg; Manganese, 520 mg; Cobalt, 50 mg; Iodine, 50 mg; Selenium, 9 mg; lasalocid, 450 mg. [2]DM = dry matter; OM = organic matter; CP = crude protein; EE = Etheral extract; NDF = neutral detergente fiber; ADF = Acid detergent fiber; TDN = Total digestible nutrients [estimated according to Cappelle et al. (2001), TDN (%) = 91.0246 – 0.571588 * NDF (diet)].

Average daily weight gain (ADG) was calculated as the difference between final live weight (FBW) and initial live weight (IBW) after the adaptation period divided by the days in feedlot receiving the final finishing diet (84 days). The weighings were carried out after fasting from water and diet for 16 hours. Dry matter intake (DMI) was estimated in eight animals from each experimental treatment (n=32), using fecal production and indigestible neutral detergent fiber (iNDF) methodologies. To estimate the fecal production (FP), titanium dioxide (TiO_2) was used as external indicator. TiO_2 was packaged in paper cartridges and introduced directly into the animal's esophagus with PVC applicator, with 10g of TiO_2 being administered to each animal for 12 days.

Fecal samples from each animal were collected daily during the last five days of indicator supply. At the end of each day of collection, the feces were dried in forced ventilation oven at 55°C for 72h and then ground in knife mill to 1mm. A composite sample from each animal was used to determine the indicator concentration. TiO_2 concentration in fecal samples was analyzed according to Detmann et al. (2012). A standard curve was prepared using 2, 4, 6, 8 and 10mg of TiO_2 and readings were taken on spectrophotometer at 410nm wavelength. To estimate FP by TiO_2, the equation was used:

$$FP = [TiO_2 \text{ supplied} / (TiO_2 \text{ in feces} / DM\ 105°C)].$$

where FP = fecal production obtained by TiO_2 concentration (g DM/day); TiO_2 supplied = amount of TiO_2 supplied to animals per day (10g); TiO_2 in feces = percentage of TiO_2 present in feces (%); DM 105°C = dry matter of feces at 105°C.

iNDF was used as internal indicator and was obtained from *in situ* incubation of fecal samples (iNDF feces) and diet (iNDF diet) for 288 hours in the rumen of a cannulated cattle receiving diet composed of corn silage and concentrate (ground corn, ground whole soybeans, urea and mineral core) (Detmann et al., 2012). To calculate DMI, the equation was used:

$$DMI = [FP \times (iNDF\ feces / iNDF\ diet)]$$

where DMI = dry matter intake; PF = fecal production; iNDF = indigestible neutral detergent fiber. Feed conversion was calculated by dividing dry matter intake by live weight gain.

Methane and nitrous oxide emissions

Feces and urine of two animals of each treatment were collected and mixed to form a composite sample. Each composite sample was applied in individual chamber, totaling 5 repetitions (chambers) per treatment (using excreta from 10 animals). Feces was collected (approximately 2 kg of feces from each animal) immediately after defecation or directly from animals' rectum. For urine collection, cattle were manually stimulated until urination and was collected 1.3 L/animal. Individual samples of excreta from each treatment were collected to determine the N concentration by Kjeldahl method (AOAC, 1995) and C concentration by dry oxidation.

In the confinement area, a pen was isolated for three months (without cattle access) before the beginning of gas monitoring (N_2O and CH_4) using closed static chamber technique.

A total of 20 plots of approximately 2 x 2 m were delimited in the pen area, in which closed static chambers (Alves et al., 2012) were centrally placed. The treatments were established by adding feces plus urine of Nellore or crossbred (Nellore x Angus) fed 65 or 85% concentrate in diet to the inner side of the chamber base. The pen area remained isolated until the end of gas monitoring to avoid external influences such as cattle trampling and deposition of excreta.

For the N_2O and CH_4 emission evaluation, the closed static chamber method was used. The chambers were produced by the researchers of the present study at Embrapa – Maize and Sorghum with a base of steel frame (60 cm length, 40 cm width, and 8 cm height) and a 1.5 cm wide U profile welded on the perimeter of the base. Thus, forming a hollow box with a trough on the upper side, which was inserted 8 cm into the soil, two weeks pretrial. Chamber height was 45 cm and the deployment time was 45 minutes (ratio of 60 cm/h) with a thermally insulated bottomless box made of PVC to avoid large differences between internal and external temperatures. A trough around frame top was filled with water at the time of gas monitoring to ensure the chamber seal.

The excreta were applied immediately after collection to the animals and only once at the beginning of winter, when cattle are usually sent to feedlots in Brazil. An amount of 1.3 kg of feces and 1.3 L of urine (weighed on digital scale) from each repetition were centrally and homogeneously added in the chambers. Gas monitoring started on the same day, and on each sampling day, gas measurements were always conducted from 09:00 h to 11:00 h, period where the measured flux is expected to represent the mean daily flux.

In the first week, the sampling frequency was daily and then, at about every three days. When a rainfall event occurred, the plots were sampled daily for three days. During chamber deployment, headspace air samples (25 mL) were taken using 60 mL polypropylene syringes at 0, 15, 30, and 45 minutes after sealing the chambers. The sample collected was transferred to previously evacuated 20 mL chromatography vials (Labco, Lampeter, U.K.).

The N_2O and CH_4 concentrations were determined by gas chromatography using Shimadzu GC-2014 (Shimadzu, Tokyo, Japan) equipped with flame ionization and electron capture detectors, back-flush and automatic gas injection system (Shimadzu, AOC-5000). The increase or decrease of N_2O and CH_4 concentration within the chamber headspace for the gas samples collected at 0, 15, 30 and 45 min were generally linear ($R^2 > 0.90$). The N_2O and CH_4 hourly fluxes (μg/m^2/h) were estimated using linear regression according to the change in gas concentration within the chamber over time (De Klein et al., 2012) following Equation:

$$F=[(\delta Gas/\delta t) \times (M/Vm) \times H]$$

where f refers to the hourly flux of N_2O or CH_4 (μg N or C/m^2/h); δGas refers to the change in headspace gas concentration of N_2O or CH_4 over time (μL/L); δt refers to the enclosure period (hours); M refers to the molar weight of N in N_2O or C in CH_4; Vm refers to the molar volume of gas (L/mol) at the temperature of the headspace during sampling; H refers to the height of headspace (mm).

Cumulative emissions from each chamber were determined as the sum of total emissions from each chamber over 84-d period, considering homogeneous fluxes. Cumulative emissions per chamber were divided by the total weight gain (E/ADG) per animal in the 84-d period, and expressed in μg/m^2/kg BW. Cumulative emissions per chamber were also divided by total dry matter intake (E/DMI) per animal over the 84-d period and expressed in μg/m^2/kg DM.

Statistical analysis

The statistical assumptions of normality and homoscedasticity were evaluated using the Shapiro-Wilk and Bartlett tests. The average performance and emissions data were subjected to analysis of variance in 2 (two breeds – crossbred or Nellore) x 2 (two nutritional levels – 65 or 85% concentrate) factorial arrangement and the means were compared using the Tukey test (P <0.05). All analyzes were performed using the R Core Team (2019) software.

Results

The initial weight, final weight and ADG were 8.92% (378 vs. 415 kg), 11.3% (534 vs. 602 kg) and 16.2% (1.86 vs. 2.22 kg/day) lower in Nellore compared to crossbred animals (P<0.05) (Table 2). The DMI was 6.28% (9.98 vs. 9.39 kg DM/animal/day) higher in animals consuming the diet with low compared to those with high nutritional level. Nitrogen content in feces was 9.7% (1.95 vs. 2.16% DM) lower in animals consuming low compared to high nutritional level diets. CH_4 emission was 2.4 times (0.662 vs. -0.916 µg C/m^2/day) higher in crossbred compared to Nellore animals (P<0.039). N_2O emission was 50.1% (228 vs. 457 µg N/m^2/day) lower in animals consuming diet with low compared to those with high nutritional level.

Table 2: Carbon and nitrogen concentrations on excreta, animal performance and intake, and daily average emissions of methane and nitrous oxide of different beef cattle breed fed different nutritional level finished in feedlot under tropical conditions

Variable	Breed		NL		SEM	P-value		
	Nellore x Angus	Nellore	L	H		B	NL	B*NL
Performance and intake								
IBW, kg	415A	378B	393	398	4.69	<0.001	0.448	0.329
FBW, kg	602A	534B	572	562	8.04	<0.001	0.363	0.446
ADG, kg/day	2.22A	1.86B	2.13	1.95	0.062	0.002	0.102	0.871
DMI, kg DM/animal/day	10.2	9.20	9.98a	9.39b	0.247	0.219	0.045	0.810
FC, kg DM/kg BW	4.65	5.01	4.77	4.89	0.140	0.661	0.215	0.417
Excreta carbon and nitrogen concentrations								
FN, % DM	1.96	1.94	1.95b	2.16a	0.042	0.119	0.005	0.184
FC, % DM	39.4	40.3	39.8	39.7	0.262	0.766	0.790	0.053
UN, g/L	18.7	20.2	19.5	21.6	0.724	0.849	0.141	0.207
Daily average emissions of methane and nitrous oxide								
ACH_4E, (µg C/m^2/day)	0.662A	-0.916B	0.368	-0.623	0.3933	0.039	0.172	0.489
AN_2OE, (µg N/m^2/day)	400	286	228b	457a	54.86	0.261	0.035	0.544

IBW = initial body weight; FBW = final body weight, ADG = average daily gain; DMI = dry matter intake; FC = feed conversion; FN = Feces nitrogen; FC = feces carbon; UN = urine nitrogen; ACH_4E = Average methane emission; AN_2OE = Average nitrous oxide emission; NL = nutritional level; L – low; H = high; SEM = standard error of mean; B = p-value for breed effect; NL = p-value for nutritional level; B*NL = p-value for interaction between breed and nutritional level effect; DM = dry matter; Means followed by different letter in the same line, uppercase comparing breed and lowercase nutritional level, differ by Tukey test.

The interaction between breed and nutritional level had significant effect between on CH_4 and N_2O E/ADG and E/DMI ($p<0.05$) (Table 3). CH_4 E/ADG of animals fed low and high nutritional levels were 2.8 and 0.95 times higher in crossbred compared to Nellore animals. Emissions in crossbred and Nellore animals were also 1.05 and 0.95 times higher when fed at low compared to high nutritional level. CH_4 E/DMI of animals fed low and high nutritional levels were 2.9 and 0.94 times higher in crossbred compared to Nellore animals. Emissions in crossbred and Nellore animals were also 1.05 and 0.84 times higher when fed at low compared to high nutritional levels.

N_2O E/ADG of animals fed high nutritional level were 18.2% (312 vs. 264 µg N/m2/kg BW) higher in crossbred compared to Nellore animals (Table 3). Emissions in crossbred and Nellore animals were also 1.22 and 1.03 times lower when fed at low nutritional levels. N_2O E/DMI of animals fed high nutritional level were 36.1% (56.5 vs. 41.5 µg N/m^2/kg BW) higher in crossbred compared with Nellore animals. Emissions in crossbred and Nellore animals were also 1.29 and 0.92 times lower when fed at low compared to high nutritional level.

Table 3: Relationship between average emissions of methane and nitrous oxide according to body weight gain and dry matter intake by different beef cattle breed fed different nutritional level finished in feedlot under tropical conditions

Nutritional level	Breed		SEM	P-value		
	Nellore x Angus	Nellore		B	NL	B*NL
Methane emission according to body weight gain (µg C/m^2/kg BW)						
Low	0.769Aa	-0.427Ba	0.0859	<0.001	<0.001	<0.001
High	-0.044Ab	-0.833Bb				

Methane emission according to dry matter intake (μg C/m^2/kg DM)						
Low	0.135Aa	-0.071Ba	0.0177	<0.001	<0.001	<0.001
High	-0.008Ab	-0.131Bb				
Nitrous oxide emission according to body weight gain (μg N/m^2/kg BW)						
Low	140Ab	130Ab	12.27	<0.001	<0.001	0.027
High	312Aa	264Ba				
Nitrous oxide emission according to dry matter intake (μg C/m^2/kg DM)						
Low	24.6Ab	21.6Ab	2.705	<0.001	<0.001	0.002
High	56.5Aa	41.5Ba				

[1]SEM = standard error of mean; [2]B = p-value for breed effect; [3]NL = p-value for nutritional level; [4]B*NL = p-value for interaction between breed and nutritional level effect; C = carbon; N = nitrogen; DM = dry matter; BW = body weight. Means followed by different uppercase letter in the same line or lowercase letter in the collum differ by Tukey test (P<0.05).

Discussion

Studies on GHG emissions from excreta from confined beef cattle have been developed in many countries, such as Australia (Bai et al., 2015; Redding et al., 2015), Canada (McGinn and Flesch, 2018; McGinn et al., 2019) and United States (Aguilar et al., 2014; Parker et al., 2018; Parker et al., 2019). However, only studies of Maciel et al. (2021) and Teixeira et al. (2023) evaluated CH_4 and N_2O emissions from excreta from confined cattle in Brazil and the present study is the first to evaluate the effect of different nutritional levels on these emissions.

Brazil is one of the countries with the largest cattle herds in the world and according to the Brazilian Association of Meat Exporting Industries (ABIEC) (2023) Brazil confined 7.62 million heads in 2022 (18% of annual slaughters), being the Nellore breed is the most used (Ferraz and Felício, 2010). Furthermore, MCTI (2020) pointed out that of the 439,213 Gg CO_2eq emitted by agriculture in 2016 (33.6% of total Brazilian emissions), 4.1% was emitted by excreta management, which shows the importance of these animals reared in this system and these emissions in the country. Another important factor is the increased grains inclusion in diets of confined beef cattle in Brazil (Silvestre and Millen, 2021) and other countries (Samuelson et al., 2016). This increase was demonstrated by Silvestre and Millen (2021), where they observed that only 58.1% of Brazilian nutritionists used more than 71% of concentrate feed in the diet of confined beef cattle in 2009, a value that reached 97.2% of nutritionists in 2019. Therefore, these data show the importance and changes that have been taking place in Brazilian beef cattle feedlots.

The greater CH_4 emission from excreta of crossbred animals compared to Nellore is a result not yet described in the literature. One of the factors that could be linked to this greater CH_4 emission is the excreta carbon concentration. According to the IPCC (2006), CH_4 emission occurs mainly in anaerobiosis, with high nutrients level for bacterial growth, at neutral pH (close to 7.0), and in warm temperatures, which indicates that environmental conditions in the present study was not entirely suitable for CH_4 production and may also justify lower or negative fluxes. However, no changes were observed in carbon content in excreta. Therefore, other factors not studied in the present study may probably be linked to this greater emission.

CH_4 fluxes varied between positive and negative values between breeds and nutritional levels, always close to zero during the evaluations. This behavior indicates that the soil acts as source and sink of CH_4 from the atmosphere at certain times. However, this pattern is inconsistent with other data available in the literature that showed short and high flux immediately after excreta application. This process occurs due to fermentation and methanogenesis still coming from the digestive tract of cattle (Angel et al., 2012; Mazzetto et al., 2014; Cardoso et al., 2016). Low or negative CH_4 fluxes after excreta application also was observed by Maciel et al. (2021) in feedlot pens. These low fluxes may have occurred due to the excreta cooling between collection from cattle and application to pens, which may reduce bacterial viability and CH_4 production. Another possible mechanism is the CH_4 loss from animal fermentation in progress in the management of excreta between collection and application.

The CH_4 net flux is regulated by processes of methanogenesis and methanotrophy (Nichols et al., 2016). Normally, CH_4 flux decreases few days after initial peak and remains at basal levels until new rain events, when new increases in fluxes occur due to anaerobic environment favoring methanogenesis. In the present study, the occurrence of CH_4 sink moments was probably due to the low rainfall volume and good soil aeration, favoring CH_4 oxidation by methanotrophs bacteria (Chen et al., 2014). Mazzetto et al. (2014) applied cattle excreta to pastures and observed summer CH_4 emissions 2.9 times higher than in winter, and that prolonged moisture conditions at high temperature resulted in higher emissions. This observation is important because most beef cattle feedlots in Brazil only operate in winter, time when there is no or little rainfall and temperatures are lower, which shows that CH_4 emissions from excreta in these systems are very low.

Furthermore, according to Liao et al. (2018) the feedlot surface with less compaction can reduce feces moisture more quickly due to the greater soil hydraulic conductivity and reduce the anaerobiosis, which justifies the low fluxes of the present study. Parker et al. (2021) also observe low CH_4 flux from feces applied in feedlot after the rain occurrence and argued that blocking the pore space in the upper feces layer can inhibit the gases diffusion from the lower layers. Furthermore, for these authors, the rainfall amount (12.7 mm) was not actually infiltrated into the feces where CH_4 is formed, which justifies the unchanged CH_4 flux.

The lower N_2O emission in animals consuming diet with low compared to those with high nutritional level is probably related to the fact that animals consuming diets with less concentrate inclusion also had lower feces nitrogen concentrations. These higher nitrogen levels in feces increase substrate availability for soil nitrification and denitrification processes, which can subsequently increase N_2O emissions (Butterbach-Bahl et al., 2013).

These higher feces nitrogen concentration may be linked to two main factors: increase in rumen undegradable protein (RUP) concentration and subclinical acidosis in animals consuming higher diet concentrate level. Animals consuming more concentrate consumed higher proportion of soybean meal and higher CP and TDN concentrations in the diet, which can increase the RUP content and nitrogen excretion in feces (Calsamiglia et al., 2010; Matthews et al., 2019). Furthermore, the greater concentrate inclusion, mainly starch, and less forage inclusion may have generated subclinical acidosis and impaired rumen fermentation (Nagaraja and Titgemeyer, 2007), reducing rumen protein use and increasing RUP. This hypothesis can be supported by the fact that animals consuming more concentrate inclusion did not have better performance.

The evaluation of diets with higher concentrate levels was carried out in the present study due to the fact that feedlots in Brazil are increasing the concentrate inclusion in diets to achieve greater performance (Silvestre and Millen, 2021). Although ruminal metabolism was not evaluated, the data obtained indicate that it is probably necessary to maximize the ruminal synchronization of carbohydrate and protein fermentation, in addition to using strategies to reduce acidosis risk, to reduce nitrogen excretion in feces and N_2O emission.

The emissions in the present study were much lower than those found by Teixeira et al. (2023) (341 µg N m^{-2} h^{-1}) and Maciel et al. (2021) (999 µg N m^{-2} h^{-1}), probably due to the lack of rainfall in the present study. This observation shows that emissions from excreta from confined cattle may be low in Brazil because most feedlots are carried out in winter. Barneze et al. (2014) and Teixeira et al. (2023) observed emission peak to occur after rainfall. Denitrification, the main process responsible for the N_2O emission, occurs mainly when the soil water-filled pore space (WFPS) is more than 70%, and there is also adequate availability of NO_3^- and carbon. Even in places where it rains in winter, constantly cleaning corrals and removing excreta before and during the rainy season can reduce N_2O emissions. In a survey carried out by Costa Junior et al. (2013) on excreta management in 73 beef cattle in Brazilian feedlots, it was observed that 48 only removed excreta at the end of confinement period and 4 did not remove waste from the pens, which shows that there is normally large excreta volume in pens and that improving the destination and treatment of this material can reduce GHG emissions.

To our knowledge, this is the first study that determined CH_4 emissions from the excreta of confined beef cattle due to weight gain and intake. The greater CH_4 E/ADG in crossbred animals compared to Nellore is linked to the higher emissions of these animals, even with the crossbred animals having higher ADG. Maciel et al. (2019) also observed greater ADG and total weight gain in crossbred animals compared to Nellore animals confined in the same location as the present study and fed 65% concentrated feed, which shows better performance of these animals in Brazil.

The higher CH_4 E/DMI of crossbred animals can also be explained by the higher emissions of these animals, since intake was equal between the breeds. These data show that crossbred animals emit more CH_4 in their excreta than Nellore animals. However, the variation between the breeds was extremely large (emission of 0.662 in crossbred and sink of – 0.916 µg C/m^2/day in the Nellore), which leads us to think that other still unknown factors, other than just the breeds, could have generated this difference. On the other hand, although daily CH_4 emissions were statistically equal, emissions from animals consuming low concentrate inclusion were numerically much higher (0.368 vs. -0.623 µg C/m^2/day), which may have generated higher emissions in animals fed low nutritional levels when dividing emissions by weight gain and intake.

The greater N_2O E/ADG in crossbred animals compared to Nellore was not expected because these animals had greater weight gain. These results are different from those found by Teixeira et al. (2023) who observed greater E/ADG in Nellore animals, but as the study by these authors did not evaluate C and N excreta concentrations, it is difficult to interpret these differences. The greater N_2O E/ADG in crossbred animals in the present study can be explained by the greater numerical flow of the crossbred animals (400 vs. 286 µg N/m^2/day), which may have generated the higher emissions when emissions were divided by weight gain and intake. The higher E/ADG and E/DMI in animals fed high compared to low nutritional levels can be explained by the higher daily fluxes in these animals.

Conclusion

Crossbred animals (Nellore x Angus) and animals consuming diets with more concentrated feed emit more CH_4 and N_2O in their excreta. These data indicate that the Nellore herd, which is predominant in Brazil, emits fewer GHGs than other breeds and that very concentrated diets have greater potential to emit GHGs.

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